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ALBERTA OIL SANDS ENVIRONMENTAL RESEARCH PROGRAM

Ecological Studies of the Aquatic Invertebrates of the Alberta Oil Sands Environmental Research Program Study Area of Northeastern Alberta

Project AF 2.8.1
April 1980

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RESEARCH REPORTS

These research reports describe the results of investigations funded under the Alberta Oil Sands Environmental Research Program, which was established by agreement between the Governments of Alberta and Canada in February 1975 (amended September 1977). This 10-year program is designed to direct and co-ordinate research projects concerned with the environmental effects of development of the Athabasca Oil Sands in Alberta.

A list of research reports published to date is included at the end of this report.

Enquiries pertaining to the Canada-Alberta Agreement or other reports in the series should be directed to:

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Ecological Studies of the Aquatic Invertebrates
of the Alberta Oil Sands Environmental Research Program
Study Area of Northeastern Alberta

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AOSERP Report 88

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The Hon. J.W. (Jack) Cookson
Minister of the Environment
222 Legislative Building
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and

The Hon. John Roberts
Minister of the Environment
Environment Canada
Ottawa, Ontario

Sirs:

Enclosed is the report "Ecological Studies of the Aquatic Invertebrates of the Alberta Oil Sands Environment Research Program Study Area of Northeastern Alberta".

This report was prepared for the Alberta Oil Sands Environmental Research Program, through its Aquatic Fauna Technical Research Committee (now the Water System), under the Canada-Alberta Agreement of February 1975 (amended September 1977).

Respectfully,

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ECOLOGICAL STUDIES OF THE AQUATIC INVERTEBRATES OF THE ALBERTA OIL SANDS ENVIRONMENTAL RESEARCH PROGRAM OF NORTHEASTERN ALBERTA

DESCRIPTIVE SUMMARY

BACKGROUND

This project, which was a continuation of 1976-77 activities of AOSERP Project AF 2.0, "Interim Report on Ecological Studies on the Benthic Invertebrates of Various Rivers in the Alberta Oil Sands Environmental Research Program Study Area, Northeastern Alberta", was to describe the basic ecology of aquatic benthic macrofauna of the Athabasca River and its major tributaries, the Muskeg and Steepbank rivers. The effect of habitat (including oil sands formations) on the diversity of the invertebrate populations and algal and bacterial responses to substrates treated with synthetic oil also were studied. Such basic taxonomic and life cycle data are required to support future manipulative and experimental studies designed to describe and predict changes in benthic communities that can be expected from oil sands development activities.

ASSESSMENT

This report has been reviewed by scientists in Alberta Environment, Environment Canada, the oil industry, and AOSERP and by a private consultant. It is the impression of AOSERP Management that a large amount of information was gathered by the study. The Alberta Oil Sands Environmental Research Program is satisfied with the efforts put forth by the researchers in this project and accepts the report, "Ecological Studies of the Aquatic Invertebrates of the Alberta Oil Sands Environmental Research Program of Northeastern Alberta", as a valid and important document suitable for wide distribution.

Wm R McDonald

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ECOLOGICAL STUDIES OF THE AQUATIC INVERTEBRATES OF THE ALBERTA OIL SANDS ENVIRONMENTAL RESEARCH PROGRAM STUDY AREA OF NORTHEASTERN ALBERTA

by

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ABSTRACT

Invertebrate fauna of the Athabasca River and its tributaries, the Muskeg and Steepbank rivers, are described qualitatively and quantitatively from baseline information gathered during 1976 and 1977. Investigations were also conducted into responses of macrobenthic communities to several potential environmental disturbances which might result from oil sands development.

Twelve sites on the Muskeg and Steepbank rivers were sampled four to five times between July 1976 and July 1977. Distinctive faunal communities reflected five principal habitats: limestone rubble, glacial till, muskeg reaches, brooks, and oil sand. More frequent collections of invertebrates from several of the same sites were used to develop more complete species lists for the area and to describe the patterns of development and life histories of some of the most common insects. Three patterns of development were recognized: fast seasonal, slow seasonal, and non-seasonal.

The general pattern of seasonal events is summarized for large tributary streams of the Athabasca River. Consequences to streams of certain oil sands development activities can be predicted. These range from total elimination of the streams to relatively minor, short-term modifications.

The Steepbank and Athabasca rivers served as a study area to assess the effects of exposure to oil sands upon composition of benthic invertebrate communities; to compare standing stocks and variety of benthos on oil sands and rubble in a riffle in the lower part of the river; and to examine the effects of a fluctuating current regime on the benthic fauna.

The upstream site had a consistently greater number of taxa. Two groups, Tanypodinae and Empididae, consistently comprised a larger fraction of the total fauna at the downstream site. The variety and density of invertebrates on oil sand were significantly less than on rubble substrates. Flooding of the riffle habitat reduced benthic standing stocks which recovered rapidly with resumption of normal current.

Summer sampling in 1977 of the Athabasca River illustrated that development of benthic communities is strongly influenced by substrate. Changes in texture of sediments and the number and variety of organisms appear to be directly linked to variations in the direction and magnitude of river currents as discharge fluctuates, and to life histories of invertebrate species.

Autumn sampling provided quantitative estimates of standing stocks of microbenthos on bedrock and macrobenthos on the entire range of sediments in the Athabasca River. The unstable sand which covers most of the river bed may prevent development of large populations of certain organisms, such as oligochetes, but does support large numbers of a few, specialized chironomids.

Studies were conducted on the responses of freshwater bacterial, algal and macroinvertebrate communities to contamination of substrates by oils. Limestone substrate contaminated with synthetic crude oil was subjected to light and dark regimes. Algal populations developed in the dark where the light level was only 0.3% at surface PAR. Oil contamination produced substantial changes in the colonization of bare stone surfaces by aquatic organisms but no great shifts in community structure occurred in response to oil contamination of established epilithic biota. Suspended and epilithic communities of the Muskeg and Steepbank rivers were found to biodegrade the saturate fraction of synthetic crude oil at 20°C and more slowly at 4°C.

Several types of material which could be used in the reclamation or diversion of streams, ranging from tailings sand to large cobbles, were compared for the nature of the macrobenthic communities which becomes established. Limestone gravel for riffles and overburden for slow reaches appear to provide for nearly natural biological productivity.

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1. GENERAL INTRODUCTION

The research detailed herein formed part of the studies in 1976-1977 on the Steepbank and Athabasca rivers of the Alberta Oil Sands Environmental Research Program (AOSERP) study area, and included a more intensive study of the Muskeg River basin.

The work included investigations on the distribution and taxonomic composition of the macrobenthic fauna of several streams and rivers in the AOSERP study area, and the responses of these communities to several of the potential environmental disturbances which might result from oil sands development.

The report is presented in three major sections. The first describes the principle habitats found in streams tributary to the Athabasca River, observations on the life histories, and some potential consequences of habitat disruption. The second section deals with the benthos of the Athabasca River and emphasizes the problems inherent in macrobenthic sampling in large rivers to detect changes in water quality. The third section describes experiments designed to assess the responses of benthic communities to specific impacts, including oil spills and substrate alterations.

Specifically, the present authors set out to gather baseline information which quantitatively and qualitatively described the invertebrate fauna of the Athabasca River, its delta, and its tributaries within the AOSERP study area throughout the year. The intent was to utilize part of this information to assess the nature and magnitude of the impact from the Great Canadian Oil Sands Ltd. (GCOS)[1] plant upon organisms inhabiting the Athabasca River. The present authors also set out to describe the responses of benthic communities of the tributary streams to such potential impacts of oil sands development as acidification, hydrocarbon seepages, spills and effluents, siltation, and stream substrate alterations.

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[1] GCOS amalgamated with Sun Oil Company on August 1979, after the writing of this report was completed, to become Suncor, Inc.

The work was done with the general intent of relating the standing stocks and population dynamics of the benthic fauna to the fish fauna of the AOSERP study area and, thereby, predict the effects of future oil sands development activities on these fauna. The studies were closely integrated with concurrent research on aquatic microbes, algae, and fisheries in an attempt to maximize the productivity of research on the basic ecology of low-gradient, muskeg-type tributary rivers and the larger Athabasca River.

2. BASELINE STUDIES OF THE MACROBENTHOS OF STREAMS TRIBUTARY TO THE ATHABASCA RIVER

2.1 INTRODUCTION

Benthic macroinvertebrates are the link between the microflora and fauna of aquatic ecosystems and higher forms, such as fish, certain mammals, and birds. While some aquatic insects are of direct economic importance to man, e.g., the blackflies or Simuliidae, the principle role of this broad group of organisms is in the transfer of nutrients both to higher forms and back to the terrestrial system via the emergence of aerial adults. Macrobenthic communities have also been shown to be sensitive indicators of present water quality and past climatic conditions (Gaufin and Tarzwell 1956; Sládecek 1973; Resh and Unzicker 1975; Clair and Paterson 1976).

In this report, investigations are described on the current distribution and taxonomic composition of the macrobenthic fauna of several streams and rivers in the AOSERP study area, as part of the efforts to obtain baseline information on the macrobenthic fauna of these rivers.

2.1.1 Methods

Twelve sites were selected to provide complete coverage of all portions of the Muskeg and Steepbank rivers (Figures 1 and 2) with respect to location within the watershed and the physical characteristics of each area. Each site was visited on 22 July and 11 October 1976, and 1 May and 16 to 22 July 1977. Sites M-1, M-2, M-4, S-6, and S-7 were also visited in late January 1977.

On each visit to each site during the open water season, 15 min were spent kick sampling (Hynes 1962a) with a coarse meshed dip net (500 µm mesh). The sampling effort was divided proportionally among the principle habitats along a 50 m reach, according to the area covered by each (i.e., riffles, pools, margins, macrophyte beds, etc.). All material collected was combined and immediately preserved in 10% formalin. An additional 10 to 15 min were spent examining

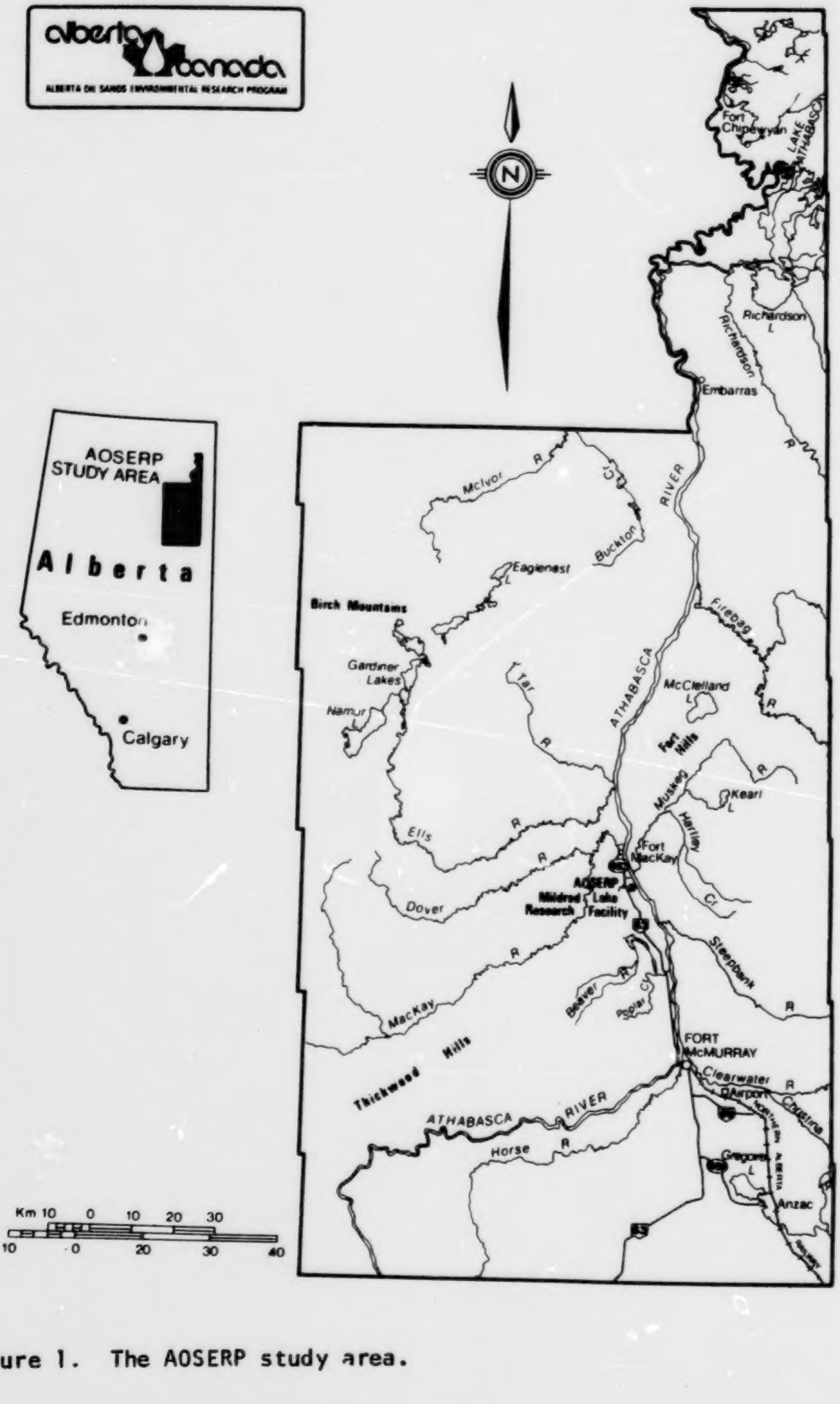

Figure 1. The AOSERP study area.

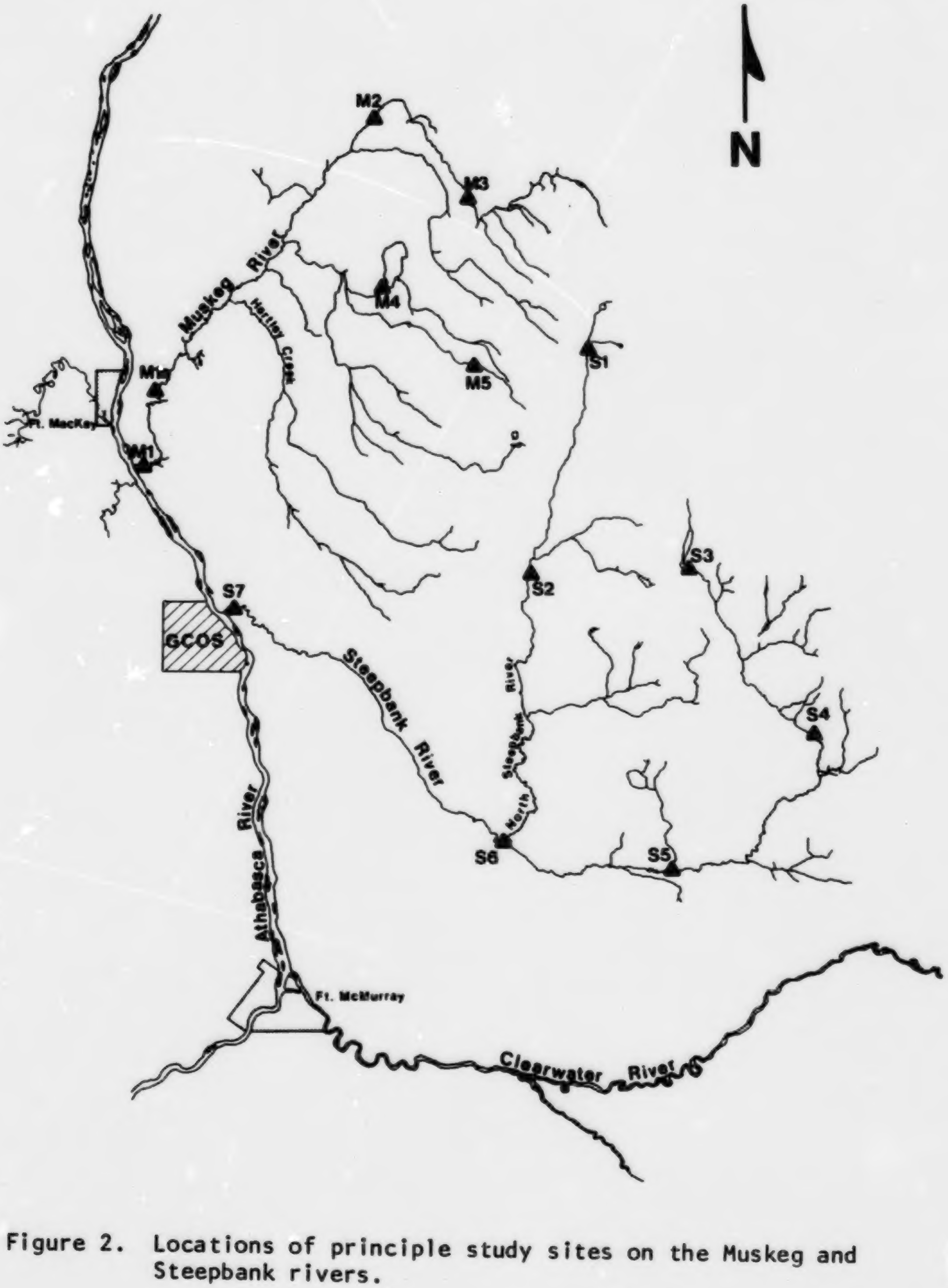

Figure 2. Locations of principle study sites on the Muskeg and Steepbank rivers.

further nettings as well as random collections of stones and debris collected by hand from which animals were picked out and preserved in 70% alcohol. Aerial adult insects were collected at each site with an aspirator, sweepnet, or by hand. In January, collecting was confined to holes cut through the ice near mid-stream.

In the laboratory, each kick sample was mixed thoroughly and a portion was withdrawn. All animals in this portion were picked from the associated debris under 10X magnification. Additional portions were examined, if necessary, until at least 300 animals had been removed. These organisms were enumerated at the generic or specific level, in most cases (see Appendix 6.1), and the percentage composition and diversity (Wilhm 1970) of the fauna were calculated. The remainder of each sample was scanned for large animals which, with those that had been handpicked in the field, were used to complete the faunal lists on a presence/absence basis.

Bray-Curtis ordinations (Bray and Curtis 1957) were performed to identify principle habitats in the Muskeg and Steepbank rivers, using all collections from each site. Two indices of similarity, Percent Similarity of Community (PSC) and Coefficient of Community (CC), were used in the ordinations:

$$PSC = \Sigma \min \left| a', b' \right| \quad \text{(Johnson and Brinkhurst 1971a)}$$

$$CC = \frac{c}{a + b - c} \quad \text{(Levandowsky 1972)}$$

Where

a' and b' = importance value (percent) of each major taxonomic group at Sites A and B, respectively,

a = number of taxa at Site A,

b = number of taxa at Site B, and

c = number of taxa common to Sites A and B.

References to types of sediments throughout this report follow the classification given by Cummins (1962).

2.1.2 Results

The general physical characteristics of each collecting site are summarized in Table 1. On the basis of these characteristics, three distinct types of stream reaches are apparent: (1) areas of swift currents and coarse sediments (gravel to boulders); (2) slow currents and fine sediments (mostly silts); and (3) sandy headwaters with moderate currents.

The mean percent composition of the animals (in major taxonomic groups) from all collections from each site (Table 2) was used to derive values of PSC (Table 3) as a basis for a Bray-Curtis ordination. This analysis generally confirmed the expected relationships between sites in terms of community composition, and suggested that S-7 was similar to, but distinct from, other type-1 habitats (Figure 3). The wide variations in the proportions of 'lower phyla', Oligochaeta, and Tanytarsini at silty, low gradient sites were reflected in the loose clustering of Sites S-1, S-3, M-2, M-3, and M-4. A second ordination (Figure 4), based upon values of CC (Table 3) derived from the lists of all taxa collected from each site, emphasized the similarities between Sites M-2, M-3, S-1, and S-3, but also indicated that Sites M-1, M-4, and S-7 were unique in terms of species composition.

On the basis of these analyses, five distinct habitats were recognized, areas with:

1. Till--swift currents (>30 $cm \cdot s^{-1}$) and substrates of coarse glacial materials (S-2, S-4, S-5, S-6);
2. Rubble--swift currents and angular limestone rubble substrates (M-1);
3. Oil Sand--swift currents over exposed oil sand (S-7);
4. Muskeg--slow currents (<10 $cm \cdot s^{-1}$) and fine sediments (M-2, M-3, M-4, S-1, S-3); and
5. Brook--moderate currents (10 to 20 $cm \cdot s^{-1}$) and predominately sandy sediments (M-5).

For convenience, these have been designated till, rubble, oil sand, muskeg, and brook, respectively.

Table 1. General characteristics of helicopter survey sites.

Site	Width(m)	Maximum Depth(m)	Mean Depth(m)	Current Speed $cm \cdot s^{-1}$	Substrate[a]
M-1	20	1.0	0.4	60	limestone rubble, gravel, boulders
M-2	8	2.5	0.8	5	silted gravel, organic debris
M-3	5	1.0	0.8	3	silt, organic debris
M-4	1-3	1.0	0.5	<1	organic debris
M-5	1	0.7	0.3	15	sand, organic debris
S-1	3	1.0	0.8	3	silted gravel, organic debris
S-2	6	1.5	0.7	30	cobbles, gravel, sand
S-3	1-3	0.6	0.3	5-20	silt, sand, cobbles
S-4	6	1.5	0.7	30	cobbles, gravel, sand
S-5	10	0.7	0.4	70	boulders, gravel
S-6	15	1.0	0.5	100	boulders, gravel
S-7	20	0.8	0.3	60	oil sand, limestone rubble

[a] Classification after Cummins (1962).

Table 2. Mean percentage composition and total number of taxa collected, July 1976 to July 1977.

Organism	M-1	M-2	M-3	M-4	M-5	S-1	S-2	S-3	S-4	S-5	S-6	S-7
Lower Phyla	3	28	34	17	18	19	5	18	4	1	2	4
Oligochaeta	4	11	27	48	12	9	3	4	4	9	5	3
Ephemeroptera	12	<1	1	<1	2	1	5	3	16	11	24	29
Odonata	1	<1	<1	<1	-	<1	<1	<1	<1	-	<1	<1
Plecoptera	4	-	-	-	3	<1	3	-	4	5	5	1
Trichoptera	7	-	2	<1	1	1	6	3	13	7	20	<1
Hemiptera	<1	2	2	<1	-	<1	<1	1	<1	-	<1	3
Coleoptera	4	-	-	<1	1	-	4	<1	<1	2	<1	<1
Simuliidae	1	-	<1	-	-	3	<1	8	1	1	1	4
Empididae	<1	-	-	-	1	<1	<1	<1	<1	1	1	7
Tanypodinae	6	2	2	4	4	7	2	2	2	1	1	4
Chironomini	4	18	12	9	7	24	3	11	<1	1	1	2
Tanytarsini	30	19	4	3	24	18	35	19	22	33	15	30
Orthocladiinae	17	15	13	14	23	7	20	10	26	17	18	9
Diamesinae	<1	<1	-	-	1	-	<1	-	-	<1	1	<1
Other Diptera	1	<1	1	1	2	1	2	2	2	9	3	1
Mollusca	6	5	3	4	<1	10	10	18	3	1	1	<1
No. of Taxa	166	81	78	87	65	103	140	119	113	105	118	81

Table 3. Values of Percent Similarity of Community (upper) based on mean percent composition and Coefficient of Community (lower) based on all taxa collected at each site.

SITES	M-1	M-2	M-3	M-4	M-5	S-1	S-2	S-3	S-4	S-5	S-6	S-7
M-1	-	52	36	38	65	52	84	56	78	81	68	69
M-2	17	-	74	59	73	78	52	70	48	48	41	43
M-3	14	43	-	75	58	58	37	56	34	34	30	29
M-4	13	30	38	-	59	54	36	49	32	33	29	28
M-5	17	21	19	11	-	67	67	65	65	65	54	54
S-1	24	36	38	35	20	-	52	76	43	43	38	45
S-2	37	25	20	16	23	31	-	60	71	77	59	60
S-3	26	37	34	25	23	38	33	-	51	46	43	51
S-4	30	25	20	14	20	25	47	30	-	71	78	62
S-5	30	15	12	11	25	17	35	25	42	-	69	61
S-6	35	16	13	10	21	20	36	27	39	40	-	62
S-7	31	17	17	9	19	23	37	24	35	41	42	-

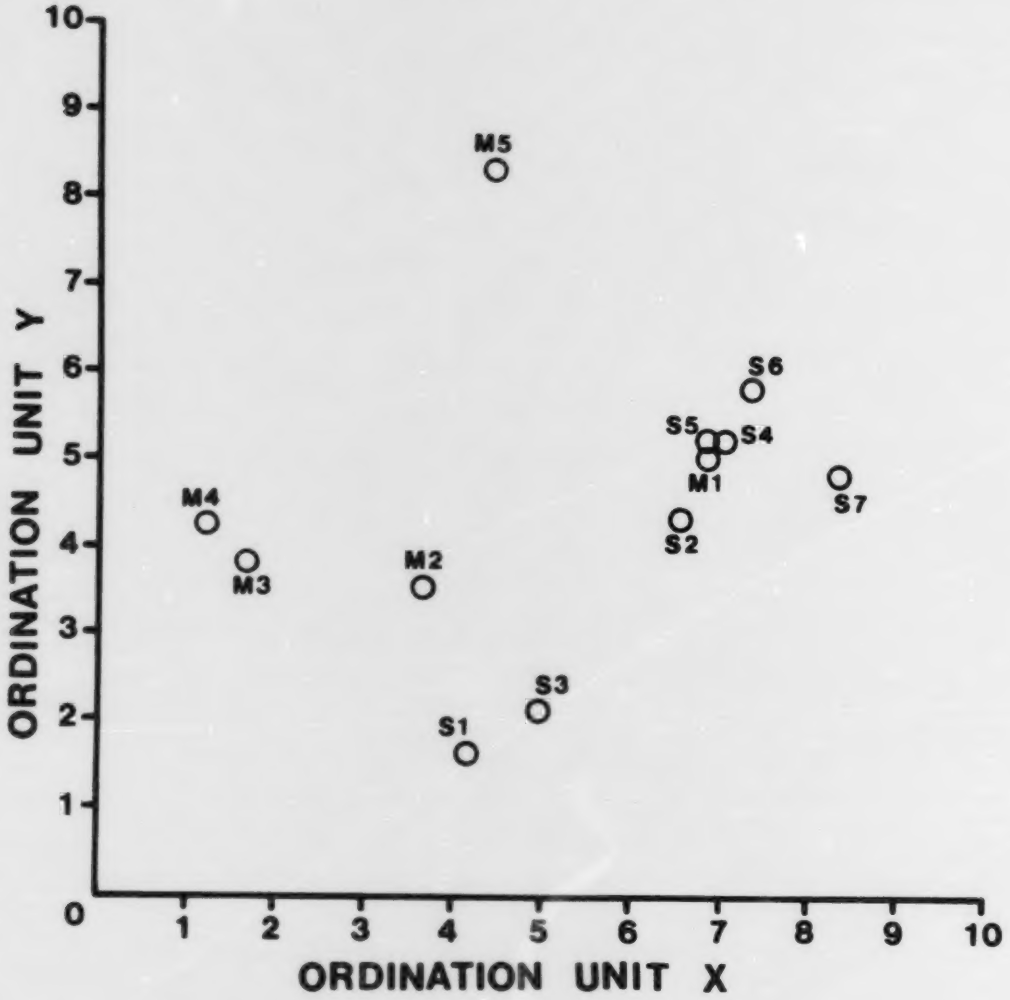

Figure 3. Ordination of survey sites based on Percent Similarity of Community (PSC). Ordination interval X corresponds to increasing coarseness of substrate and Y corresponds to other factors including current speed and detritus.

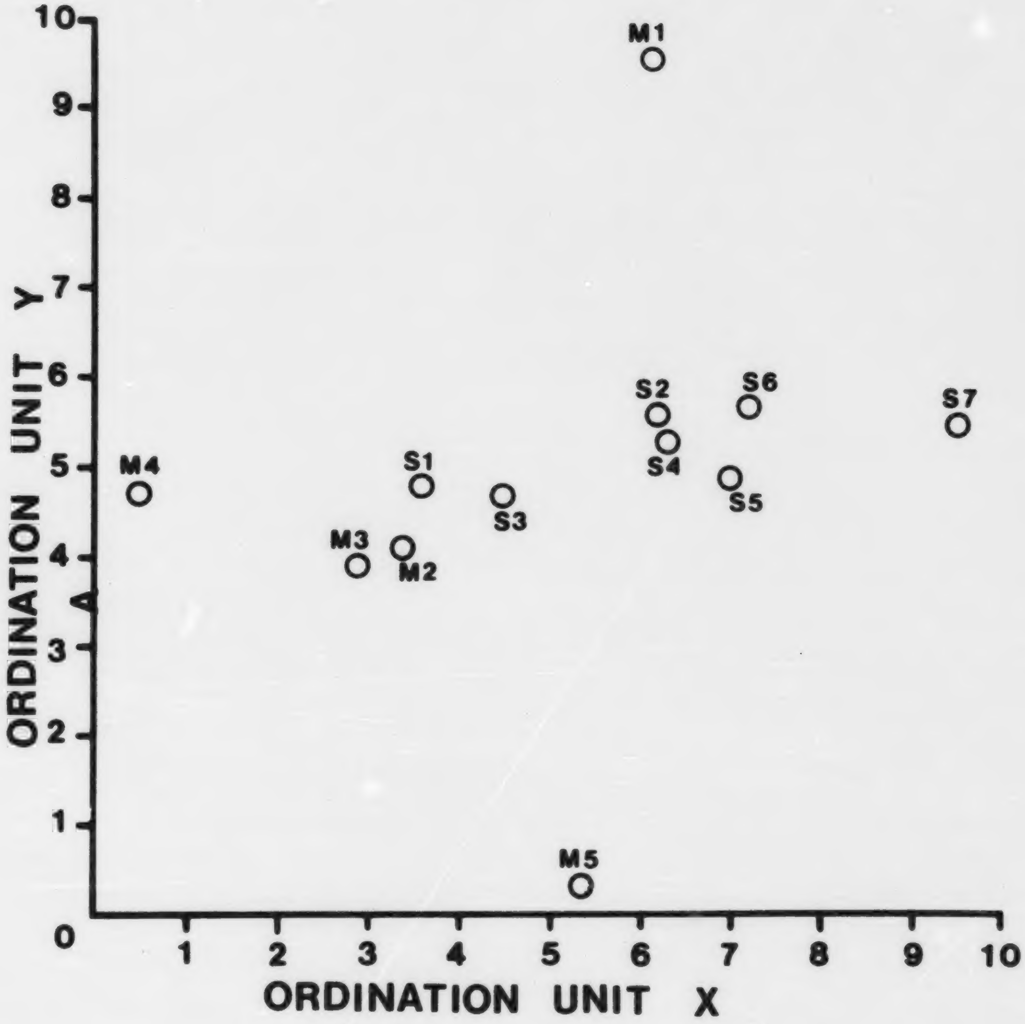

Figure 4. Ordination of survey sites based on Coefficient of Community (CC). Interval X corresponds to increasing coarseness of substrate and Y corresponds to other factors including current speed and detritus.

On the basis of species composition, M-4, the outlet of Kearl Lake, could be separated from the other muskeg sites but, because of its general similarity in percent composition, this has not been done. Similarly, the rubble habitat at M-1 was combined with till habitats for the consideration of general community structure.

The mean percent compositions of all collections from each of the principle habitats on each collecting date were used to calculate PSC between successive sampling visits and between years (Table 4). These values indicated that the communities of muskeg and rubble/till habitats were the most consistent between years, and that of the brook site, the least. Community composition tended to be most stable during the winter except at S-7.

While the relative proportions of the most abundant groups of organisms varied between visits and between sites, certain groups characterized the fauna of each habitat. A variety of species of Tanytarsini, Ephemeroptera, Trichoptera, and Orthocladiinae were dominant at rubble and till sites. The most abundant animals on oil sand over the entire study were *Baetis* sp. (Ephemeroptera) and Tanytarsini (mostly *Rheotanytarsus*). The muskeg fauna was composed mainly of Oligochaeta, Copepoda, Chironomini, and species of Orthocladiinae usually not abundant on rubble and till. The brook fauna was dominated by *Stempellina* (Tanytarsini) and a third assemblage of Orthocladiinae. Species unique to M-5 included *Trichotanypus posticalis* and *Protanypus* sp.

Diversity ($\bar{d}$) generally differed little between seasons and was greatest in collections from rubble/till habitats (Table 5). The low diversity on oil sand in January, as well as the apparently great dissimilarity between this collection and those of October and May (Table 4), probably reflected the very limited area which could be sampled due to the thickness of the ice in mid-winter. Rubble/till also yielded the greatest number of taxa per date and per site. The mean number of taxa per site ranged from one third to one half of the total number of taxa per date at rubble/till

Table 4. Percent similarity of community between successive sampling visits.

	HABITAT			
Date	Rubble/till	Oil Sand	Muskeg	Brook
July 1977				
	72	57	77	29
July 1976				
	68	40	59	50
October 1976				
	82	23	67	
January 1977				NS[a]
	59	41	56	
May 1977				
	62	78	67	48
July 1977				

[a] NS = not sampled.

Table 5. Number of taxa and diversity ($\bar{d}$) in collections from principle habitats.

	Rubble/Till	Oil Sand	Muskeg	Brook
July 1976				
Total number of Taxa	140	20	92	17
Mean Taxa/Site	50		33	
$\bar{d}$	4.336	2.890	3.620	3.508
October 1976				
Total number of Taxa	118	38	94	31
Mean Taxa/Site	50		32	
$\bar{d}$	4.379	4.367	3.492	3.763
January 1977				
Total number of Taxa	77	14	28	
Mean Taxa/Site	49		16	NS
$\bar{d}$	4.172	1.787	2.968	
May 1977				
Total number of Taxa	132	39	99	28
Mean Taxa/Site	55		38	
$\bar{d}$	3.951	3.596	3.589	3.937
July 1977				
Total number of Taxa	163	43	107	28
Mean Taxa/Site	62		42	
$\bar{d}$	4.398	3.618	3.634	3.446
Total number taxa (all collections)	262	83	213	66

and muskeg sites. The analysis of larger portions of each sample might have increased these fractions somewhat, but rare taxa, occurring as one or two specimens per sample, would probably still have accounted for a large number of all taxa collected on any given date.

2.1.3 Biology of selected invertebrates

The life cycles and general biology of some of the more abundant or unusual aquatic invertebrates were studied in detail to provide a more complete picture of the aquatic fauna of the AOSERP study area. The information presented in this section is based upon the material collected during the helicopter survey and other individual studies described elsewhere in the report section, as well as additional kick samples, random collections, and observations made throughout the field seasons, especially at Site M-1A (Figure 1). Except where otherwise noted, insufficient specimens were collected from any one site or experiment to elucidate the life history of any species, so material from all sources has been combined to present the general growth patterns of various organisms. These general patterns can be expected to be obtained over most of the study area, but specific details of the life history and behaviour of individual species will probably vary from place to place and from year to year. In several cases, obvious differences between habitats have been noted.

2.1.3.1 Oligochaeta. Oligochaeta were identified using keys and descriptions found in Brinkhurst and Jamieson (1971), Brinkhurst (1964), and Kennedy (1969), and by comparison with previously confirmed specimens in the author's own collection. *Nais behningi* was the most abundantly collected species and occurred with *Nais simplex*, *Pristina breviseta*, and Enchytraeidae in areas of moderate to swift current in the Muskeg, Steepbank, and Athabasca rivers. Other Naididae were usually found among aquatic macrophytes and in muskeg reaches. Tubificidae were most abundant in muskeg habitats and on mud in the Athabasca River. Other species of this family undoubtedly occur in the study area, but sexually mature worms were rarely collected.

2.1.3.2 Lower Taxa. This diverse, artificial grouping used for convenience throughout this report includes Porifera, Cnidaria, Turbellaria, Nematoda, Nematomorpha, Hirudinoidea, Mollusca, Tardigrada, and Crustacea, except where otherwise noted. The pooling of these obviously unrelated groups is justified by the fact that, with certain exceptions, none were taken abundantly in terms of numbers or biomass. It is interesting to note that Clifford (1969) found Ostracoda to be a numerically dominant group in the Bigoray River, the only muskeg stream which has been extensively studied in North America. While often encountered in the collections of this study, ostracods were seldom abundant. Whether this difference is real or merely a reflection of annual variations or sampling effort cannot be assessed. The amphipods, *Gammarus lacustris* and *Hyalella azteca*, listed as commonly collected and locally abundant in the Muskeg and Steepbank rivers, were found only in quiet water-backwaters, among macrophytes, and in ponds and lakes.

2.1.3.3 Ephemeroptera. The 37 Ephemeroptera listed in Appendix 6.1 do not constitute a complete list for the AOSERP study area, but probably include most of the common, and many rare, lotic species or genera. Species determinations have been based on adults, or for certain well-known groups such as *Ephemerella*, mature nymphs. Two poorly known, but very important, genera, *Baetis* and *Heptagenia*, are each represented by at least three species in the study area, but further study is needed to separate them.

All three patterns of development for aquatic insects (fast seasonal, slow seasonal, and non-seasonal) described by Hynes (1970) were found among the Ephemeroptera. *Ephemera simulans* and *Hexagenia* probably require two years to complete development from egg to adult (Coleman and Hynes 1970; Barton 1976) and thus represent the non-seasonal pattern. Both are restricted in their distribution: *E. simulans* was found frequently only in the Muskeg River above Site 1A, where the current is not turbulent and the substrate consists of limestone bedrock with a thick layer of aufwuchs and some silt. This species also occurs with *Hexagenia* in the Athabasca delta and at least a few of the deeper lakes in the area.

Fast seasonal or summer species include *Siphlonurus alternatus* (Figure 5), *Analetris eximia* (Lehmkuhl 1976), *Metretopus borealis*, *Cloeon* sp. (Clifford 1969), *Paraleptophlebia* sp. (Figure 6), *Tricorythodes minutus* (Figure 7), *Brachycercus* sp. (Figure 8), and at least two species of *Heptagenia*. These mayflies overwinter as diapausing eggs, hatch in late May, develop rapidly, and emerge and reproduce in July or early August. With the exception of *A. eximia*, *Paraleptophlebia* sp., and *Heptagenia* spp., the preferred habitat of mature nymphs of each of these species is quiet backwaters with abundant vegetation or organic debris. *Paraleptophlebia* sp. and *Heptagenia* spp. were found in gravelly or rocky riffles. *Analetris eximia* was found only in the Athabasca River, a significant range for this endangered species, formerly known only from portions of the Black Forks (Wyoming) and Saskatchewan rivers (Edmunds and Koss 1972; Lehmkuhl 1976).

The emergence period of *T. minutus* (Figure 7) is quite long and it is possible that more than one generation is produced during the summer. The life history of *Metrotopus borealis* remains somewhat in doubt, due to the small number of collections in which this species appeared, but seems to conform to the fast seasonal pattern. Nymphs 7 mm in length were collected in early June. Growth was rapid through mid-July, and mature male nymphs were 1 to 1.5 mm shorter than females. Development of a population in a cold brook near Site S-7 appeared to be delayed about two weeks, in comparison with that in the Muskeg River.

The eggs of slow seasonal mayflies hatch shortly after being laid and the young nymphs grow rapidly until water temperatures fall in autumn. The nymphs grow slowly in winter and then rapidly complete their development and emerge in spring. This pattern appears to apply to *Ameletus* sp.; young and half-grown nymphs were collected in mid-October. Mature female nymphs, in very early May,

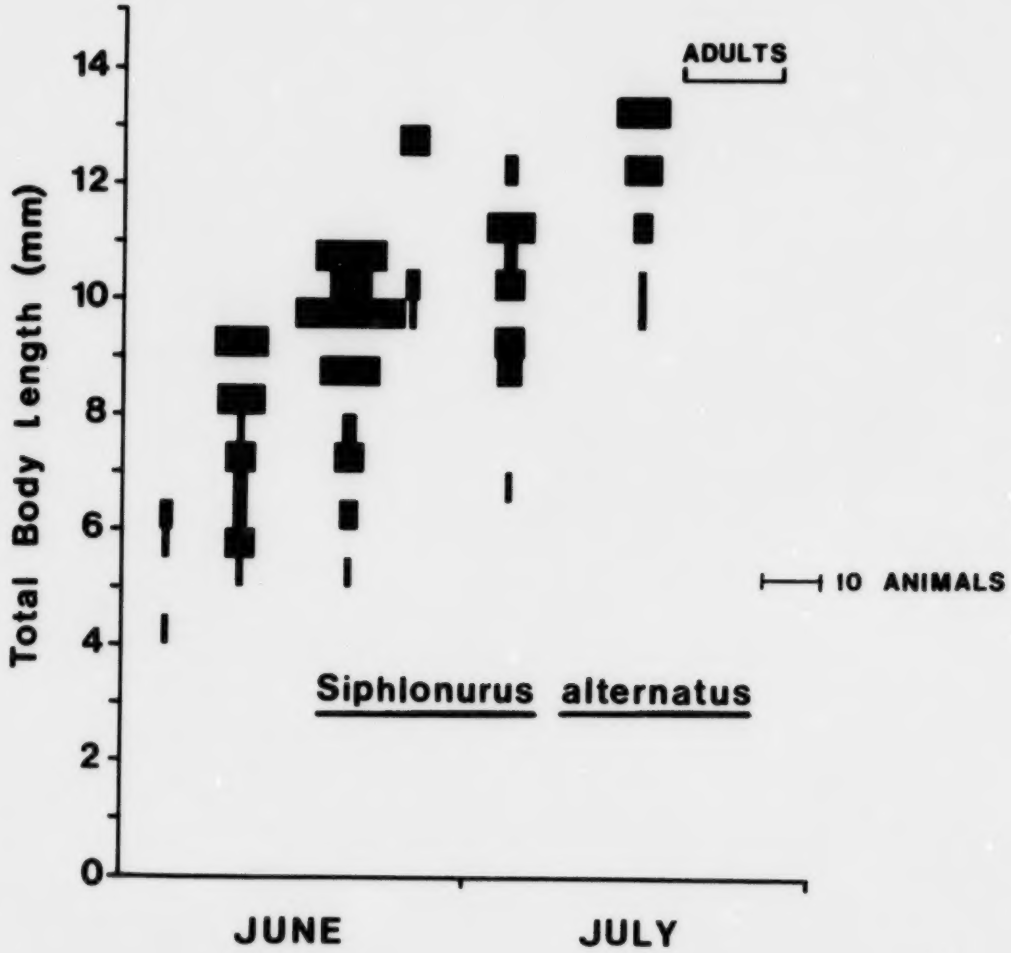

Figure 5. Numbers of *Siphlonurus alternatus* in 0.5 mm size classes, 1976-1977.

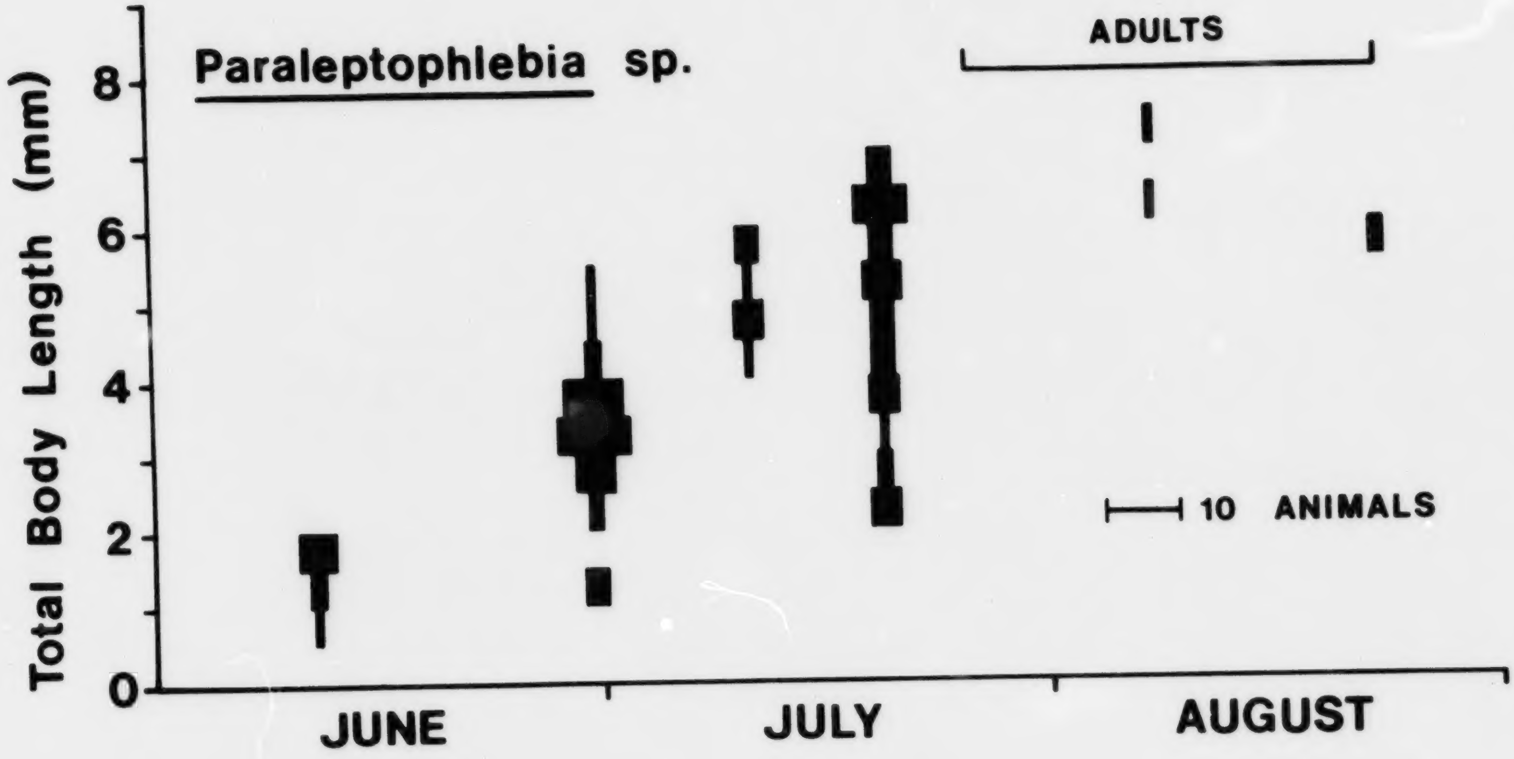

Figure 6. Numbers of *Paraleptophlebia* sp. in 0.5 mm size classes, 1976-1977.

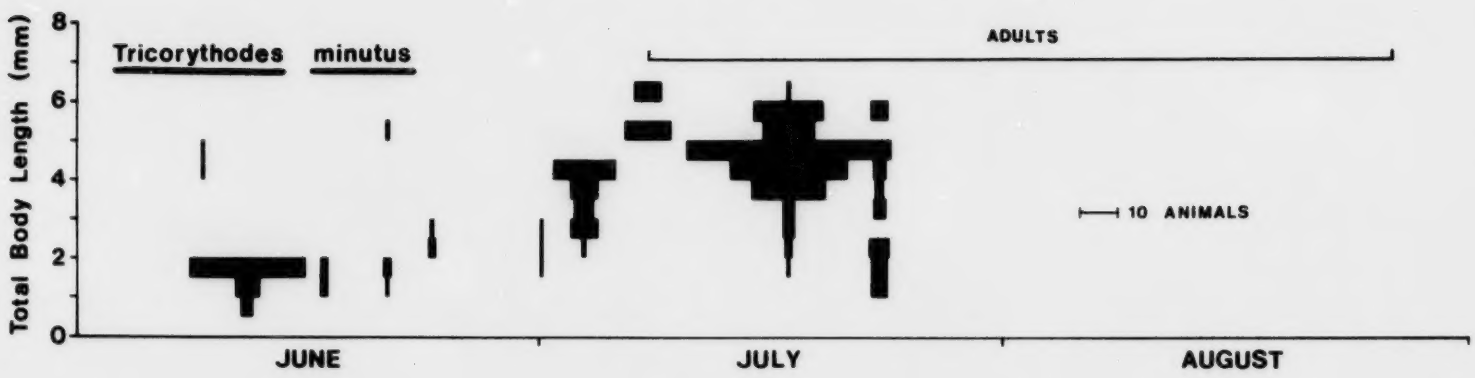

Figure 7. Numbers of *Tricorythodes minutus* in 0.5 mm size classes, 1977.

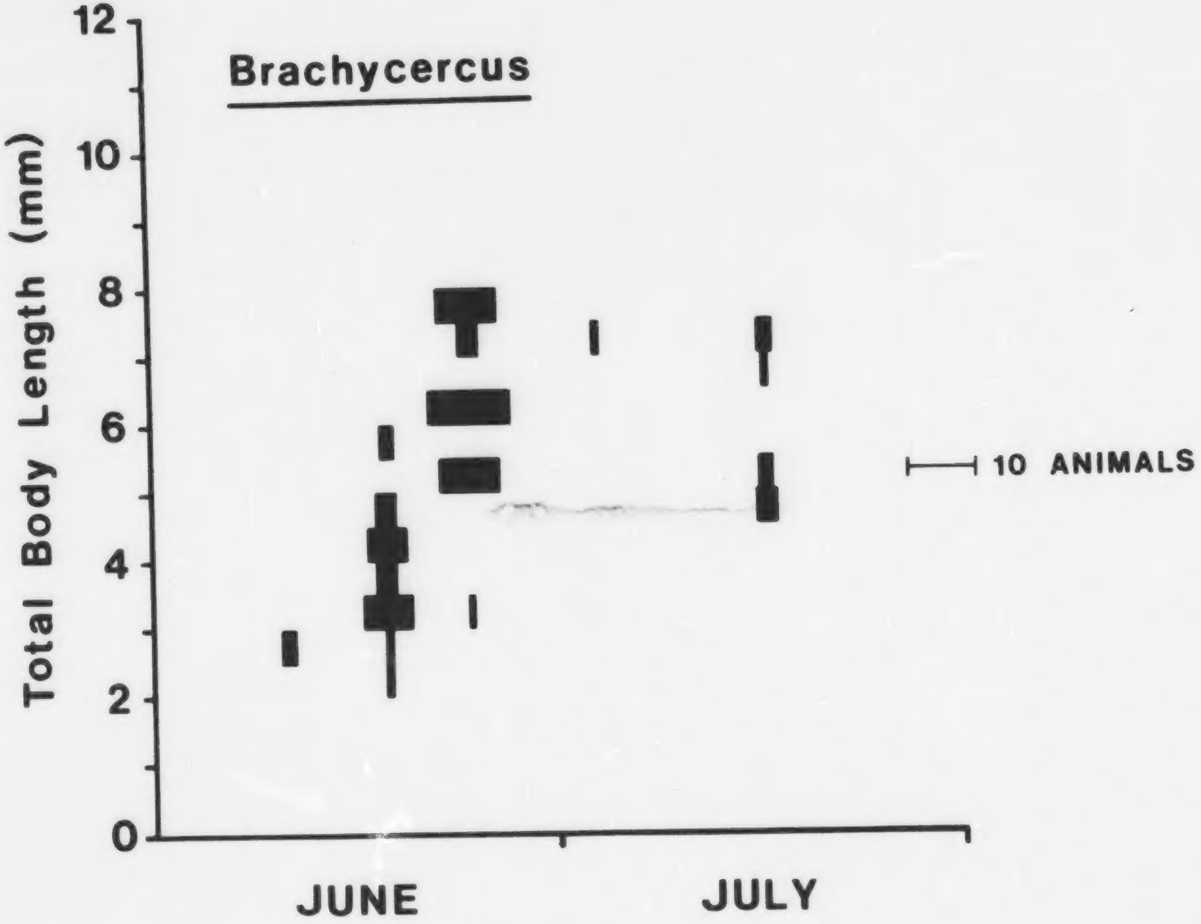

Figure 8. Numbers of *Brachycercus* in 0.5 mm size classes, 1976-1977.

were 1.3 mm longer than males. The life history of *Leptophlebia cupida* in the Muskeg River followed the pattern described by Clifford (1969), including migration into tributary streams as the ice broke up in April (Hayden and Clifford 1974).

With the possible exception of *Baetis*, the most abundant mayfly in riffles of the lower Muskeg River and on bedrock in the Athabasca River was *Ephemerella inermis* (Figure 9). This species is very tolerant of silty water (Allen and Edmunds 1965), but was abundant only in non-depositional areas. *Ephemerella inermis* has been reported to be multivoltine in some locations (Allen and Edmunds 1965), but is univoltine and slow seasonal in northern Alberta. Adults emerge from early June through late July, and there is an extended hatching period from July through October. Growth is rapid through autumn, slow in winter, and rapid again in May.

Ephemerella spinifera was rarely collected in the Muskeg River, but was abundant on moss-covered stones in riffles and rapids in the Steepbank River. Separation of nymphs of this species from those of the closely related and often sympatric *E. grandis* is difficult, but the size of mature nymphs corresponds to measurements of *E. spinifera* given by Allen and Edmunds (1962a). No adults were taken, but emergence probably occurs in June. Young nymphs were abundant in mid-July and were half-grown by mid-October (Figure 10). There appeared to be very little growth in winter.

Both of these *Ephemerella* species are predominately western in their distributions (Allen and Edmunds 1962a, 1965). Among the other species of the genus found in the study area, *E. simplex* has previously been reported only as far west as Manitoba, *E. tibialis* is a western species, and disjunct eastern and western populations of *E. aurivilli* and *E. margarita* have been reported (Allen and Edmunds 1961, 1962b, 1963, 1965).

The genera *Baetisca, Stenacron,* and *Stenonema* in North America are principally eastern in their distribution. One species, *B. columbiana,* found in the Steepbank and Muskeg rivers, appears to have been reported only as the single nymph from the Columbia River,

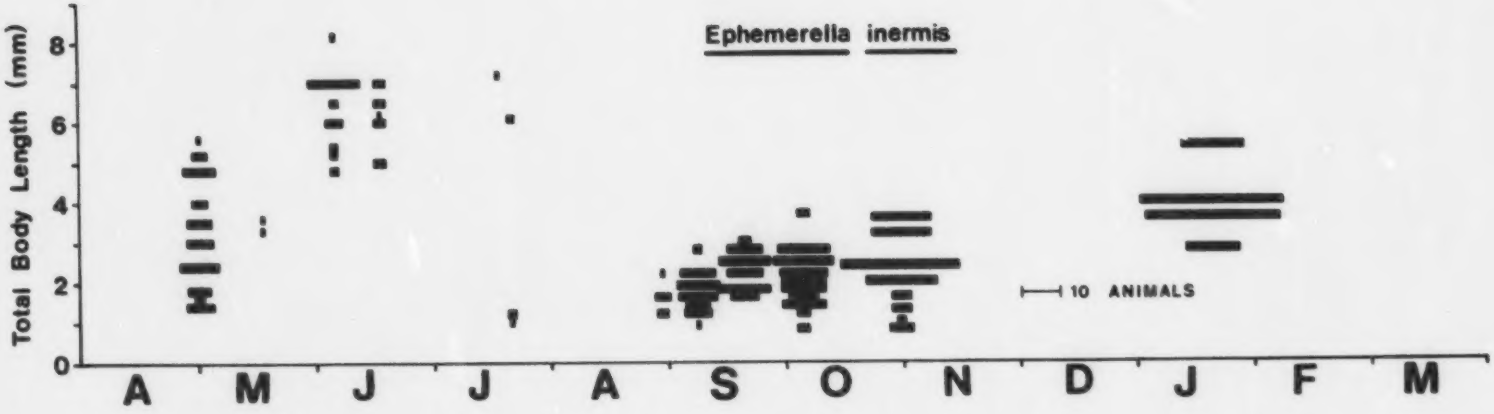

Figure 9. Numbers of *Ephemerella inermis* in 0.2 mm size classes, 1976-1977.

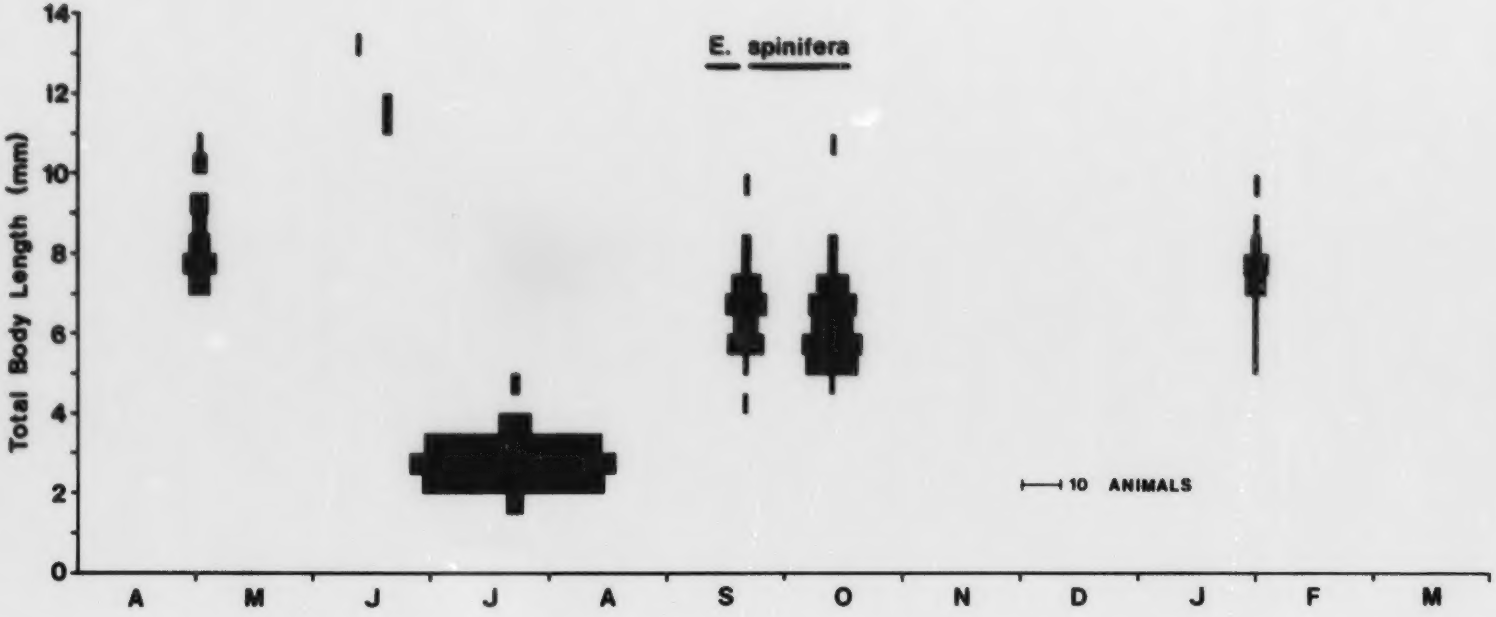

Figure 10. Numbers of *Ephemerella spinifera* in 0.5 mm size classes, 1976-1977.

Washington, U.S.A., from which the species was described (Edmunds 1960). Only one (undescribed) species of *Stenonema* has been recorded west of Ontario (Edmunds et al. 1976). *Baetisca obesa* seems to be widespread in the Mackenzie River drainage (D. Rosenberg, Freshwater Institute, Winnipeg, Manitoba). Since nymphs of *B. obesa, Stenacron interpunctatum*, and *Stenonema vicarium* are all known to inhabit both rivers and the wave-washed shores of large lakes (McDunnough 1933; Edmunds et al. 1976; Barton and Hynes in press), it is probable that postglacial dispersal from the Mississippi River basin through the Laurentian Great Lakes into Lake Agassiz, enabled these species to reach the Athabasca River (Elson 1967; Lehmkhul 1972).

2.1.3.4 Odonata. Only two species of dragonfly nymphs were commonly collected from swiftly flowing streams in the study area. Both were abundant but rarely were found together. Large populations of *Somatochlora minor* were found on moss-covered stones in riffles in the upper and middle Steepbank River and the lower reaches of the Pierre and Calumet rivers. Adults were collected in June. Walker and Corbet (1975) described this as a species preferring small, clear, gently flowing streams, a description which fits the Calumet River very well.

Walker (1958) described *Ophiogomphus colubrinus* as a northern species inhabiting clear, rapid streams with gravelly or sandy beds. Nymphs of this species were abundant in riffles in the lower Muskeg River (rubble habitat) and common in the lower Steepbank River and on bedrock in the Athabasca River. This species has a very long flight period; adults were collected or observed from 9 June through 30 September 1977. Since females oviposit over much of the summer, distinct cohorts of nymphs were not distinguishable. A complete range of sizes of nymphs was collected throughout the year suggesting that this species has a life cycle of at least two years.

Nymphs of the other species of Odonata were found in more lentic habitats. *Gomphus ?notatus* was collected only in muddy backeddies of the Athabasca River. The aeshnids, cordulids, and agrionids were most commonly found in slowly flowing muskeg habitats or in weedy backwaters in the lower Muskeg and Steepbank rivers. Species of *Leucorrhinia* and *Libellula* were usually found on vegetation in pools of muskeg terrain. All of these species were on the wing in mid-June.

2.1.3.5 Plecoptera. The 21 species of stoneflies found in the study area also included fast, slow, and non-seasonal species. Fast seasonal species included the winter stoneflies *Capnia vernalis*, *Oenopteryx fosketti*, *Taeniopteryx nivalis* (Figure 11), *T. parvula*, and perhaps *Nemoura arctica* and *N. rotunda* (Figure 12). Adults of *Capnia* and the Taeniopterygidae emerge as the ice goes out in April. Eggs hatch about one month later (Taeniopterygidae) and the young nymphs undergo diapause during the summer (Harper and Hynes 1970), or the eggs remain in diapause until August (*Capnia*). Emergence occurred in May (*rotunda*) or late May and early June (*arctica*) in *Nemoura*. Growth of all species was rapid through autumn and winter. Many *T. nivalis* had already reached full size by late January.

Slow seasonal development was observed in *Arcynopteryx* sp., nymphs of which were fully grown in October (Figure 13), and four other species. Eggs of *Isogenus frontalis colubrinus* hatched from June through August. Nymphs grew rapidly through autumn (Figure 14), but there was a wider range of sizes of nymphs in the Athabasca than in the tributary streams. Peak emergence probably occurred in May and adults were collected as late as early July.

Both adult emergence and egg hatching were prolonged over several months in *Hastaperla brevis* (Figure 15). Growth was slow through autumn and winter and rapid in spring. This species was very abundant in riffles or tributary streams and on bedrock in the Athabasca River, spreading onto gravel and coarse sand as discharge declined in autumn.

The two abundant species of *Isoperla*, *I. ?fusca* and *I. longiseta*, were restricted to tributary streams and the Athabasca River, respectively, and differed somewhat in their life histories.

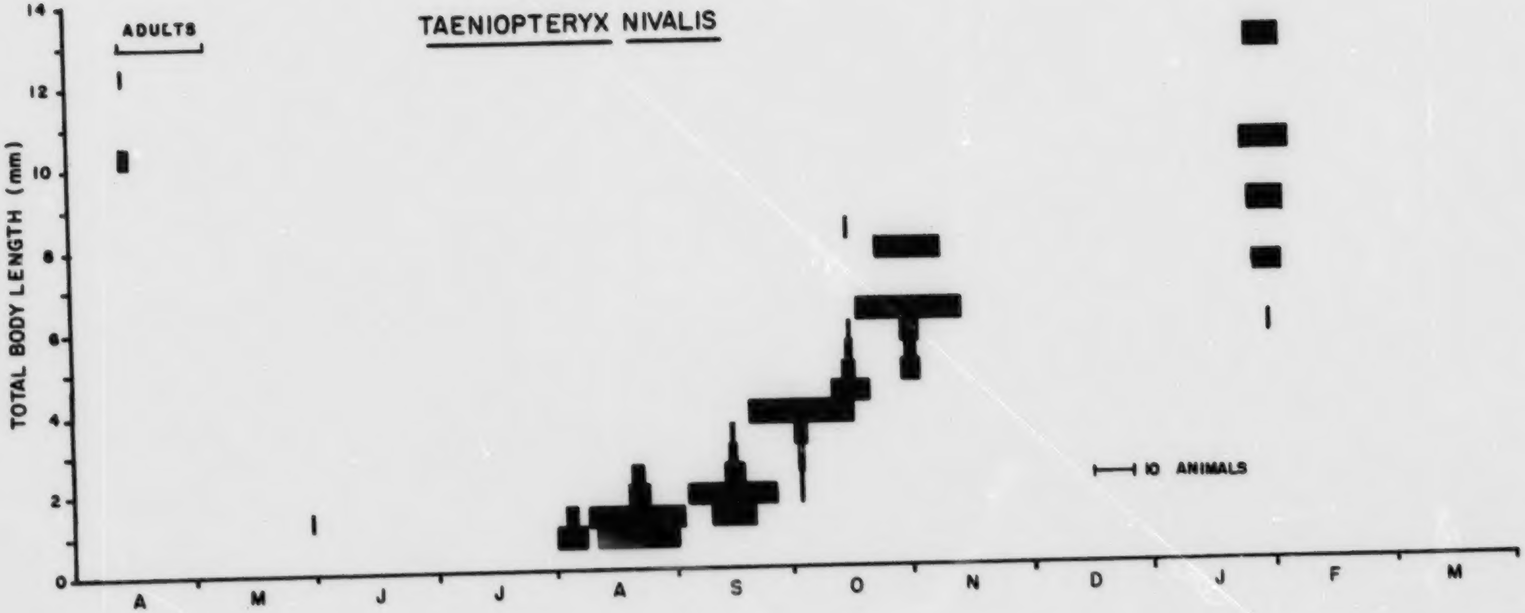

Figure 11. Numbers of *Taeniopteryx nivalis* in 0.5 mm size classes, 1976-1977.

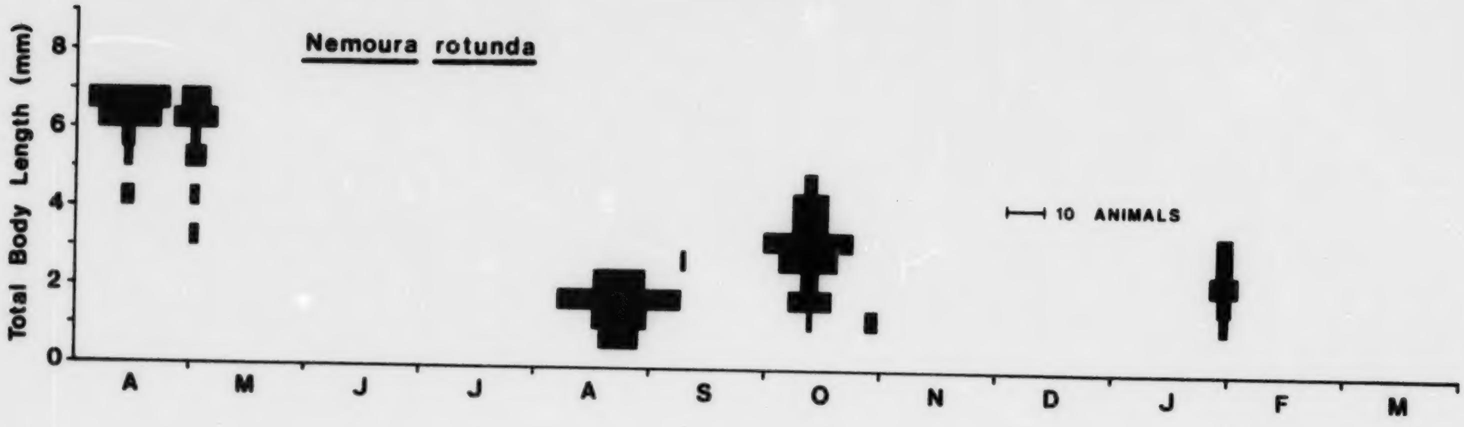

Figure 12. Numbers of *Nemoura rotunda* in 0.5 mm size classes, 1976-1977.

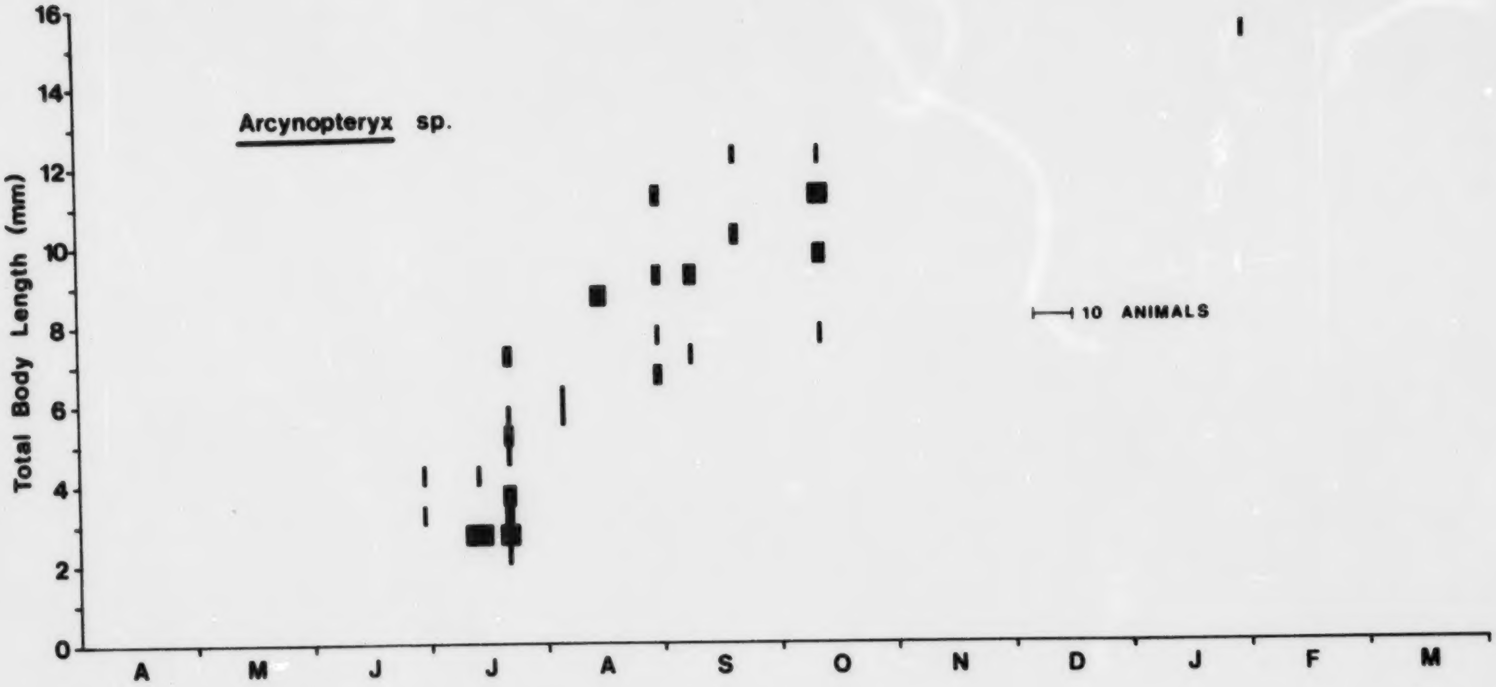

Figure 13. Numbers of *Arcynopteryx* sp. in 0.5 mm size classes, 1976-1977.

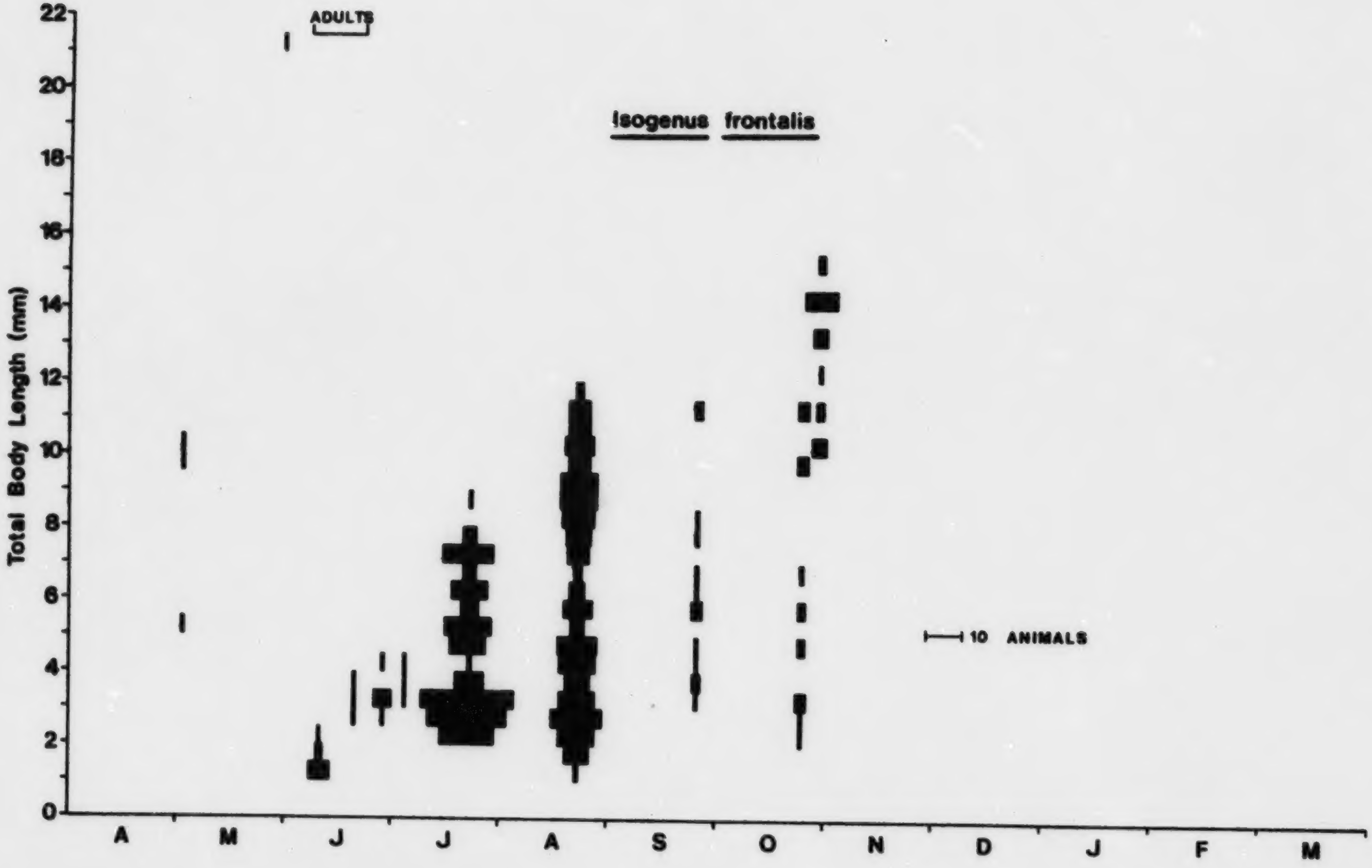

Figure 14. Numbers of *Isogenus frontalis colubrinus* in 0.5 mm size classes, 1976-1977.

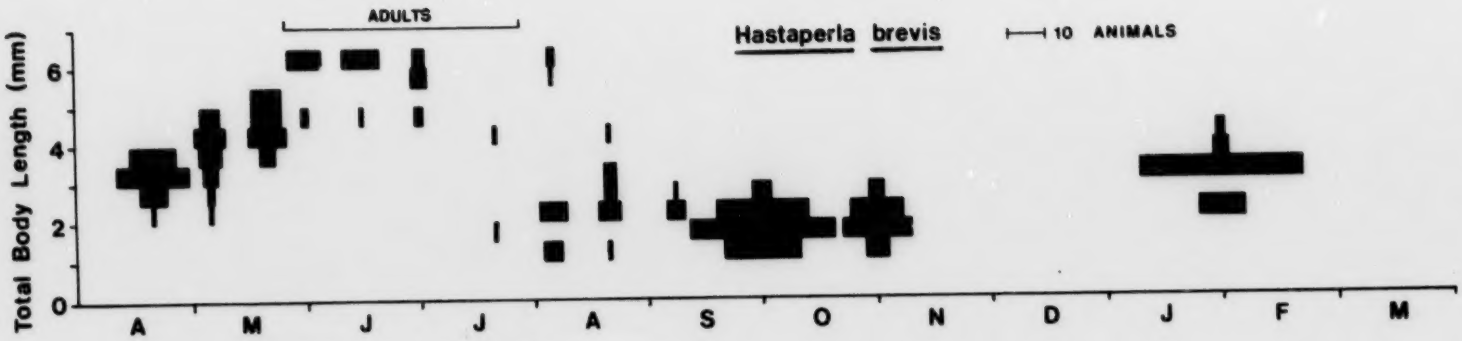

Figure 15. Numbers of *Hastaperla brevis* in 0.5 mm size classes, 1976-1977.

Adults of *I. ?fusca* were collected from late May to late June, and young nymphs appeared in July (Figure 16). Growth was slow through the following April and rapid in May. The lack of winter collections from the Athabasca River hinders the interpretation of the life history of *I. longiseta*, but it appeared that eggs hatched either in August, followed by steady growth through the winter, or in late winter and early spring, followed by rapid growth through June and July (Figure 17). Maximum emergence was observed in early June and adults were taken in lesser numbers through late August. No change was observed in the size of adults as the summer progressed. The preferred habitat of both species was similar to that of *H. brevis*, but mature nymphs of *I, longiseta* were especially abundant in accumulations of debris in the current during June.

At least four non-seasonal species required three or more years to complete their development: *Pteronarcys dorsata* (Figure 18), *Claassenia sabulosa* (Figure 19), *Acroneuris lycorias*, and *A. abnormis* (Hitchcock 1974; Flannagan 1977; Gaufin et al. 1972). A fifth species, *Pteronarcella regularis*, probably has a two-year life cycle (Gaufin et al. 1972) and was found only among debris in the Athabasca River and beneath undercut banks in the Steepbank River. *Acroneuria lycorias* was collected in small numbers in riffles and rapids in tributary streams (Muskeg, Steepbank, MacKay rivers). Adults were reared and collected in the field in mid-June. Nymphs of *A. abnormis* were found on bedrock and debris in the Athabasca River, also usually in small numbers.

Our observations indicated that *Pteronarcys dorsata* has an egg diapause of 10 to 11 months and a total of three, or in some cases four, years are required to complete the life cycle (Figure 18), a pattern similar to that described for *P. proteus* (Holdsworth 1941). Adults emerged over a very short period in the second half of May and young nymphs first appeared in the following April. Growth was fairly uniform and rapid through the second summer, but erratic in the third; some nymphs apparently reached full size 28 months after the eggs were laid, while others apparently needed another summer to

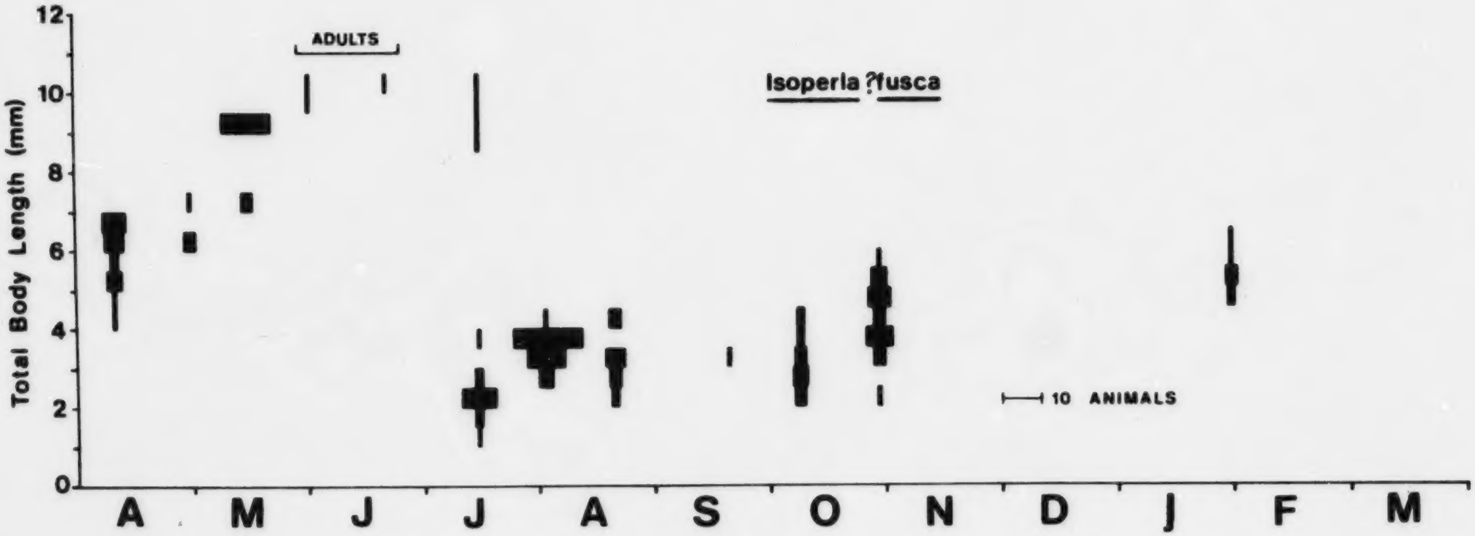

Figure 16. Numbers of *Isoperla ?fusca* in 0.5 mm size classes, 1976-1977.

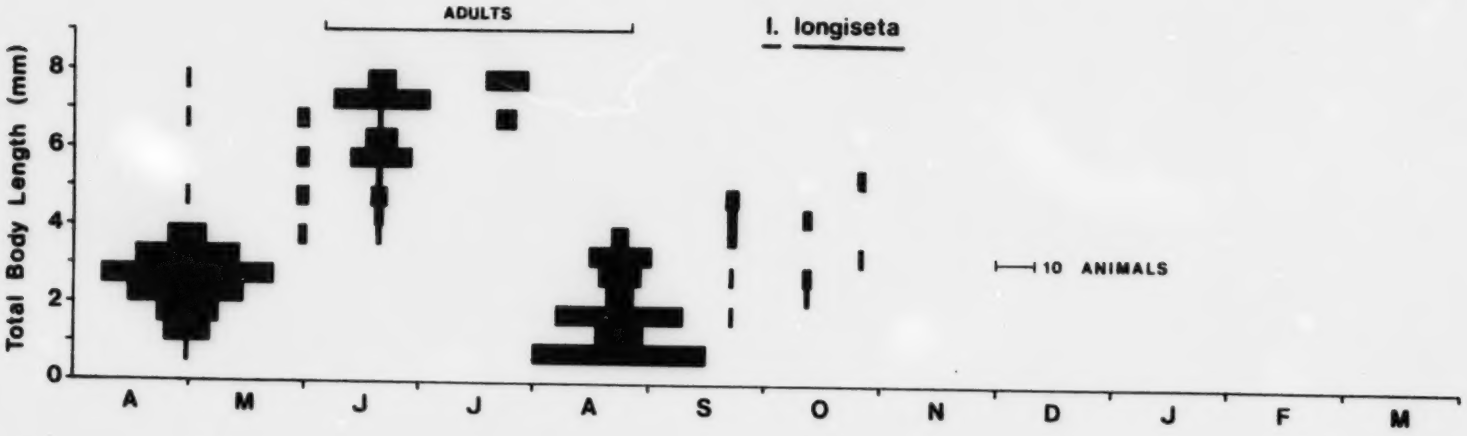

Figure 17. Numbers of *Isoperla longiseta* in 0.5 mm size classes, Athabasca River, 1977.

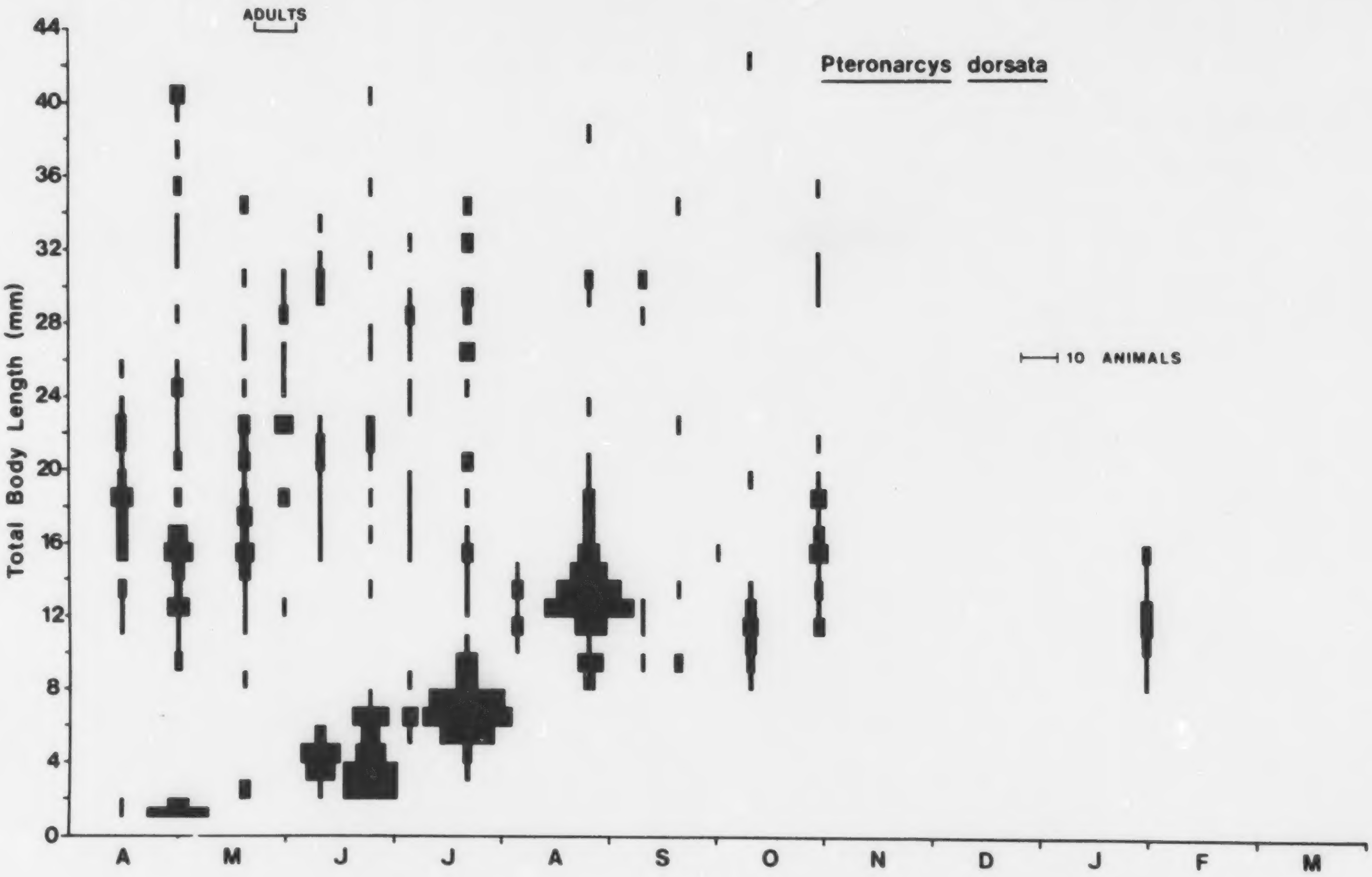

Figure 18. Numbers of *Pteronarcys dorsata* in 0.5 mm size classes, 1976-1977.

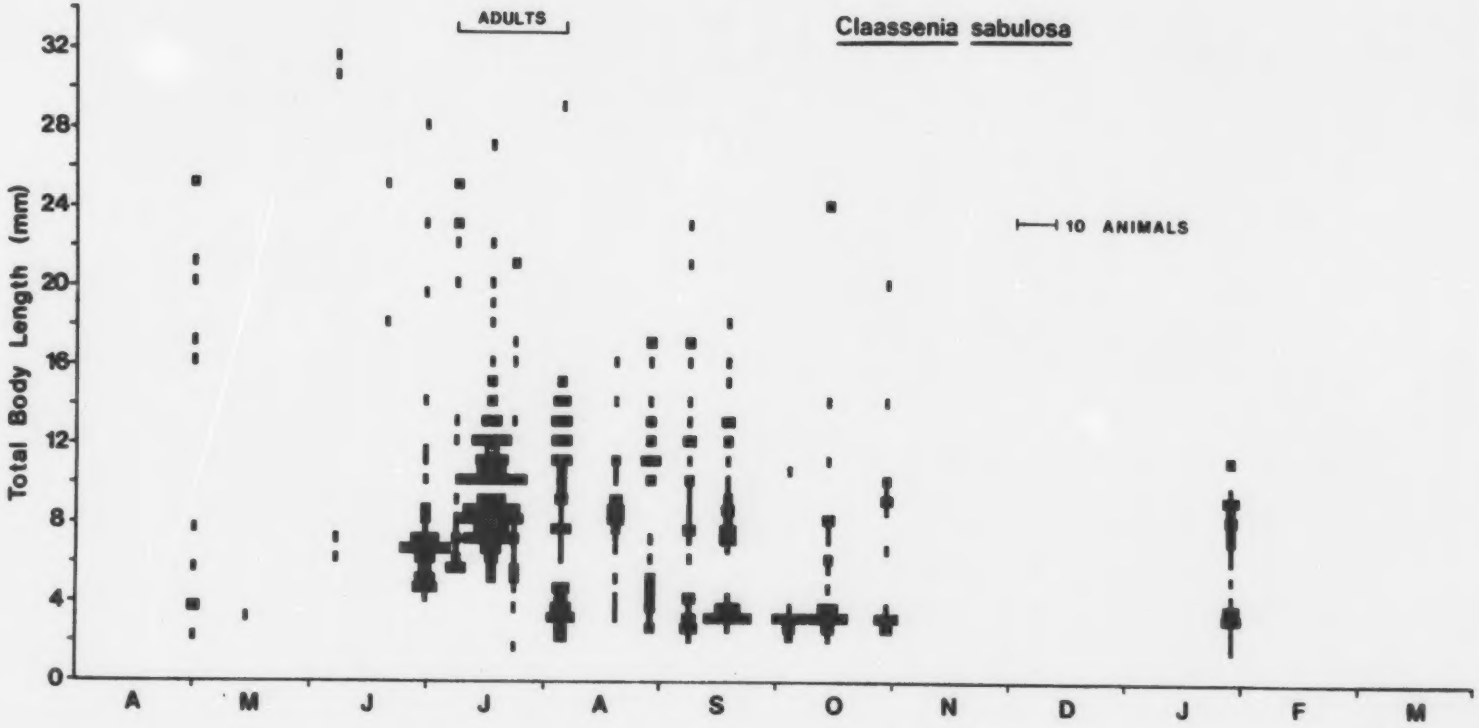

Figure 19. Numbers of *Claassenia sabulosa* in 0.5 mm size classes, 1976-1977.

complete their growth. There was little growth in winter. Nymphs were most abundant among accumulations of debris (sticks, leaves, etc.) during the summer months and probably play a major role in the secondary processing of allochthonous organic material (Short and Maslin 1977). In late summer and early autumn, the nymphs migrated to the deepest parts of riffles and rapids where they appeared to remain quiescent under large stones through the winter. Nymphs dislodged from the substrate in summer made no attempt to swim, but held all legs and antennae close to the body and drifted passively.

The observations of *Claassenia sabulosa* emphasize the hazards inherent in drawing conclusions about the biology of stream benthos based upon short-term studies. This species was very abundant in riffles in the Muskeg and Steepbank rivers in 1976, but was essentially absent in 1977. The reason for this disappearance remains a complete mystery. The 1976 collections suggested at least a three-year life cycle (Figure 19) with emergence in June and July when adults were found under stones within a few metres of the stream. Eggs appeared to hatch throughout the year.

The stonefly fauna of the AOSERP study area includes species whose principle distributions are eastern, western, and northern. The first group includes *N. rotunda* (also known from the Mackenzie Basin in the Yukon and Northwest Territories, Wiens et al. 1975), *C. vernalis, T. nivalis, T. parvula,* and both species of *Acroneuria* (Hitchcock 1974; Ricker 1946; Harper and Hynes 1971a, b). Western species include *I. fulva, I. fusca, I. sordida, P. regularis,* and *C. sabulosa* (Gaufin et al. 1975; Jewett 1959). Widespread, essentially northern species include *N. arctica, L. sara, P. dorsata, I. frontalis colubrinus, I. longiseta,* and *H. brevis* (Hitchcock 1974; Harper and Hynes 1971c; Jewett 1959; Gaufin et al. 1972; Ricker and Scudder 1975; Ricker 1946).

2.1.3.6 Trichoptera. A detailed study of caddisflies in Hartley Creek is already being carried out by R.A. Crowther (University of Calgary, Calgary, Alberta; AOSERP Project AF 2.5.1) and so less time was into the study of this group. Species names have been applied only to adult specimens; thus, several of the listed genera (Appendix 6.1) actually contain two or more species. Most adults were taken in June and July but *Psychoglypha subborealis* and *Glyphopsyche irrorata* were collected in late April and early May. *Hydropsyche bifida* and *Oecetis avara* were on the wing from June through early October. The importance of this group, especially in 'till' habitats, cannot be overstressed: while Tanytarsini were more numerous, caddisfly larvae were dominant in terms of biomass at S-2, S-4, S-5, and S-6 throughout the study period.

2.1.3.7 Hemiptera. From late summer through spring, adult corixids were found in the Athabasca River and its tributaries, where this group is an important source of food for several of the dominant fish species, especially goldeye (Donald and Kooyman 1977; Bond and Berry 1977) and lake whitefish (Bond and Berry 1977; McCart et al. 1977). With the exception of *Trichocorixa verticalis interiores* which overwinters in the egg stage in saline lakes (Tones 1977), corixids overwinter as adults (Hungerford 1948). Observations indicate that reproduction occurs only in essentially lentic habitats--lakes, ponds, muskeg pools, and backwaters, or very slow reaches of tributary streams. The new generation of adults migrate from these areas in late summer and overwinter in deep pools in streams and the Athabasca River. Deeper lakes are probably inhabited year-round. Large numbers of adults were occasionally attracted to lights in spring and late summer, suggesting aerial migrations between breeding and overwintering sites.

All of the species found in 1976-1977 have been previously recorded from northern Alberta (Hungerford 1948; Brooks and Kelton 1967). Further collecting, particularly in lentic habitats, would probably reveal several additional species.

2.1.3.8 Coleoptera. The list of beetles given in Appendix 6.1 is far from complete, except for the Elmidae. *Optioservus fastiditus* was the most commonly collected benthic species, occurring abundantly in riffles and on bedrock in the Athabasca River. *Dubiraphia robusta* was infrequently collected, usually on sandy substrates, and has previously been reported only from a lake in northern Wisconsin (W. Hilsenhoff, pers. comm.). Larvae of *Brychius* were abundant at Sites S-4 and S-5. Other species and families were typically found in lentic habitats.

2.1.3.9 Diptera. The most important groups of Diptera in terms of numerical abundance were the Tipulidae (especially *Dicranota* and *Eriocera*), Simuliidae, Chironomidae, Rhagionidae, and Empididae. *Dicranota* and *Eriocera* were abundant from autumn through spring in gravelly riffles and, as noted by Clifford (1969), grew through the winter. Detailed studies of the Simuliidae (and the other Diptera) were hampered by the poor state of taxonomic knowledge about this group. Large populations developed in early spring (*Simulium vittatum*), July (*S. arcticum*), September (*S. decorum*), or periodically throughout the openwater season (*S. tuberosum* complex). Estimates of the densities of several populations are given in succeeding sections of this report.

Larvae of the rhagionid, *Atherix pachypus*, were abundant on rocky substrates throughout the study area. They are predaceous, feeding on practically all aquatic invertebrates, and perhaps exhibiting cannibalism (Hagatomi 1962; Neveu 1976). The adults of various species feed on nectar or blood of amphibians and mammals (Nagatomi 1962), but specific information is lacking for *A. pachypus*. The two-year larval period is followed by pupation just above the water level under stones or in debris. There appeared to be little growth in winter. The larvae of Empididae are also generally thought to be predaceous (Wirth and Stone 1971), but have rarely been collected in large numbers (J.A. Downes, Biosystematics Research Institute, Ottawa, Ontario). Since it was impossible to separate larvae to species, little can be said about life histories except that development takes less than one year (some species may be multivoltine) and at least some species grow throughout the winter.

Adults of the genera and species found in the AOSERP study area have been reported to prey on blackflies (Peterson and Davies 1960; Downes and Smith 1969). Larvae of *Rhamphomyia* were not recognized in the collections, but the adults were frequently observed swarming over the lower Muskeg and Steepbank rivers.

This study of the Chironomidae must be considered as preliminary in nature, as this was the most abundant group of aquatic insects. In a detailed study of a portion of the Bigoray River, Boerger (cited in Clifford in press) found 109 species; therefore, the list of 108 taxa found during the present study must be considered far from complete. At least four previously undescribed species were found, of which three were abundant. *Rheocricotopus* nr. *kenorensis* was found in stony riffles in the Muskeg River. Third instar larvae of 'Orthocladiinae A' were extremely abundant in gravel in the lower Muskeg in the autumn of 1976. Observations on 'Orthocladiinae B' are detailed in Section 3.

The summary of the distributions of chironomids given in Appendix 6.1 may give the impression that most are generally distributed throughout the study area, but in most cases, these have been identified only to the generic level. Individual species are probably more restricted in their habitat preferences (Beck 1977).

2.1.4 Discussion

Although the word 'habitat' has been used as a generic term for the different reaches of stream recognized on the basis of the present survey of the Steepbank and Muskeg rivers, it should be emphasized that each is in fact a mosaic of microhabitats which are inhabited by various species. Thus, even the most superficially uniform habitat, 'muskeg', consists of mud, sand, organic debris, and macrophytes. The most heterogeneous habitat, 'rubble', consists of all of the former as well as particles of clean-swept angular limestone ranging in size from boulders to gravels. The physical complexity of each habitat type is reflected in the mean number of taxa collected per site per visit. Since no species was actually restricted to any one site or habitat, and the time period covered by this study was too short to permit an adequate assessment of annual variations in the relative abundance of species, each habitat was named to reflect dominant or unique physical features rather than indicator species. The fauna are also summarized in terms of the relative numerical abundance of major groups throughout the year. It should be emphasized that the dominant groups would be somewhat different if the analysis had been based on biomass rather than numbers, but the latter was chosen since both vary seasonally and numbers are easier to obtain, a factor which can be of great importance in the design of biological monitoring programs.

Having defined the principal communities of aquatic invertebrates in the Muskeg and Steepbank rivers, the life histories of some of the most common insects were considered. These were divided into three types: fast seasonal, slow seasonal, and non-seasonal. Fast seasonal species develop only at either high or low temperatures and avoid unfavourable conditions through diapause as eggs or specialized nymphs. Nymphs of most fast seasonal, or summer, mayflies tended to prefer silty backwaters where ice freezes into the substrate in winter. Fast seasonal, or winter, stoneflies avoid high summer temperatures and are consequently confined to riffles which do not freeze solidly in winter. As would be expected, slow and non-seasonal species were also most abundant in habitats which were most stable throughout the year, i.e., riffles and deep pools. As noted above, the preferred microhabitats of certain species, e.g., *Leptophlebia* spp., *Pteronarcys dorsata,* and *Taeniopteryx* spp., changed seasonally and there is evidence to suggest that much of the summer fauna of riffles in smaller streams may overwinter in deep pools to avoid being frozen (R.A. Crowther, Department of Biology, University of Calgary, Calgary, Alberta).

Using information of a similar, but much more detailed, nature drawn from 10 years of investigations on the Bigoray River in west-central Alberta, Clifford (in press) has described the normal occurrence of seasonal events in a stream similar to those in the AOSERP study area. Since the portion of the Bigoray River upon which this description is based is similar to parts of the Muskeg River, the general pattern of seasonal events can be expected to apply to all the habitats here except, perhaps, brooks which freeze completely in winter. These events can be briefly summarized for large tributary streams of the Athabasca River.

1. Winter (November to mid-April). Discharge is minimal but fairly stable beneath almost total ice cover. Temperatures remain near 0°C, but winter stoneflies, certain empidids, tipulids, and chironomids grow rapidly in riffles. Very large populations of *Simulium vittatum* develop in late winter.
2. Spring (mid-April to May). Discharge increases to maximum levels flushing fine sediments from muskeg reaches, then declines as water temperatures increase rapidly. Many stoneflies, chironomids, and several mayflies and caddisflies emerge from riffles and deep pools. Arctic grayling, suckers, and northern pike enter the streams to spawn.
3. Summer (June to August). Water temperatures increase slowly until August and discharge stabilizes except after exceptionally heavy or prolonged rain. Adult suckers migrate out of the stream; adult and young-of-the-year grayling feed heavily on aquatic and terrestrial insects. Summer and slow seasonal species complete their development and begin new generations. Aquatic macrophytes grow in most habitats.
4. Autumn (September to October). Water temperatures decline and the flow peaks briefly when summer foliage of the muskeg vegetation is killed by frost. Most fish leave the streams. Leaf fall provides maximum annual input of allochthonous organic matter which is processed by rapidly growing fast seasonal, winter species of invertebrates. The growth rates of most slow seasonal species decline.

It is clear that processes important to the functioning of lotic ecosystems in the study area occur throughout the year. The overall rate of these processes is greatest during the open water season but nutrient cycling through the winter is also crucial to the whole system.

From this general sequence of events and more specific observations, it is possible to predict some of the consequences to streams of certain activities which may be expected to accompany oil sands development. These range from total elimination of the streams to relatively minor, short-term modifications. Inclusion of a stream in a surface mining area, as has happened in the former middle reaches of the Beaver River, is, of course, the first extreme.

Less drastic, but still potentially severe, consequences can be predicted for the remaining portions of diverted streams. Judging from the existing and proposed locations of surface mining developments, two new streams will be created by the diversion of existing rivers: the former lower reaches containing far smaller volumes of water (if not completely dry), and the channels carrying and receiving water diverted from the upper reaches. Reduced discharge in the lower reaches would result in a physically smaller stream. If flow remains sufficient to avoid summer stagnation and excessively high temperatures due to the relative decrease in shading from streamside vegetation (at least until formerly inundated areas develop forest cover), summer species of invertebrates should persist and may support small fish populations during the open water season. Most, if not all, slow seasonal and winter species would be eliminated by freezing of the substrate in winter. The elimination of these species can be expected to have an effect on the remaining fauna through the system, but as yet, we have insufficient data to predict the nature of this effect or the consequences of the elimination of processed allochthonous material from muskegs upstream. Increased sediment inputs from construction activities would also be deleterious to the remaining flora and fauna (Rosenberg and Snow 1975) and this would be magnified by decreased flushing due to low flows.

The biota of the diversion and receiving channels will, of course, depend on the nature of the channels, especially with respect to gradient, substrate, bank vegetation, and the actual amount of water which is diverted, both absolutely and seasonally. If the combination of these factors approximates the situation in natural streams in the area, a normal fauna should develop within a few years. Again, the data needed to predict the time involved is lacking, but there is evidence that a conditioning period is necessary before newly inundated sediments become true streambeds (see Section 3) and several years may be required for the immigration and re-establishment of plant and animal species. If the combination of physical features is otherwise, other aquatic communities can be expected. For example, since large quantities of sand become available after the vegetation and surficial soil are stripped away, a channel could be envisioned which is basically sand, but stabilized somewhat with a layer of stones. Sand is a very poor substrate for aquatic organisms, especially in a lotic system (Nuttal 1972; Leudtke and Brusven 1976), since it does not provide firm attachment surfaces or interstitial spaces for the accumulation of organic matter and refugia for invertebrates. Therefore, the benthic fauna of such a channel would be essentially restricted to organisms which live on the upper surfaces of stones, namely Simuliidae, Hydropsychide, certain Baetidae, and perhaps a few oligochaetes and water mites. If the channel was shallow, extensive freezing of the substrate in winter would eliminate the slow seasonal species, leaving the fast seasonal simulids. Very high temperatures in summer, resulting from an absence of shading vegetation, would have a similar effect.

Clearly, this would not be a desirable situation. More productive diversion channels would resemble natural streams, consisting of long, deep (1 m or more) pools connected by riffles. A layer of gravel, cobbles, and boulders at least 30 cm thick would stabilize the riffles and support a diverse fauna. The sandy beds of the pools would soon accumulate organic material and become typical 'muskeg' reaches if at least a border of natural vegetation were left along the channel. The roots of this surrounding vegetation would also help to stabilize the banks of the channel. If such channels emptied into the original lower reaches of the stream, the natural 'rubble' and 'till' habitats, which may be vital to important fish species for spawning and feeding, would maintain nearly normal regimes of flow and nutrient inputs.

Vegetation in the watershed, and especially muskeg, has a profound effect on the hydrology of streams (Holecek and Noujaim 1975; Likens et al. 1977). Clearing of land associated with both surface mining and in situ recovery of oil from the oil sands can be expected to alter the flow regimes and nutrient balances of streams in the area. Specific, immediate biological effects will include reductions in the abundance of the mayflies *Leptophlebia cupida* and *L. nebulosa* through the elimination of emergence sites (Hayden and Clifford 1974), elimination of organisms entering the streams from muskeg pools, which are important food items for young fish (Clifford 1972), and elimination of important breeding habitats for Corixidae. These aquatic invertebrates are a major food source for several of the most economically valuable fish species in the Athabasca River, especially in late summer and autumn when individuals of other food organisms such as Ephemeroptera and Plecoptera are small and not readily available. Observations suggest that corixids do not reproduce in the Athabasca River, but rather in backwaters of tributary streams and in the numerous pools found in muskeg terrain. Adults do not seem to overwinter in the latter habitat, but migrate to larger streams and the Athabasca River. Further research is needed to assess the production of corixids in muskeg pools and the distances covered by migrating adults.

These, then, are some of the major physical disturbances which will accompany oil sands development and their biological consequences. Other potential environmental impacts are beyond the scope of this study (e.g., acid precipitation), will be considered in Section 4 (oil spills), have been discussed in the literature (e.g., siltation [Rosenberg and Snow 1975; Chutter 1969]; saline discharge--see review by Machniak 1977), or remain to be studied (e.g., the effects of clearing and draining large areas on the hydrology and nutrient balance of streams).

2.2 THE EFFECTS OF EXPOSURE TO OIL SAND ON MACROBENTHIC COMMUNITIES

Contamination of aquatic habitats resulting from accidental or experimental spills of crude and refined oils has been shown to cause changes in both the density and variety of various components of the flora and fauna (McCauley 1966; Roeder et al. 1975; Rosenberg and Wiens 1976; Parker et al. 1976; Burk 1977; Busdosh and Atlas 1977). In the Athabasca Oil Sands area of northeastern Alberta, many streams are naturally exposed to hydrocarbons as they cut through the oil sands deposit. The substrate of the lower reaches of the Steepbank River and similar nearby streams consists of patches or even continuous layers of oil sand and, thus, offered an opportunity to study benthic invertebrate communities which have been exposed to hydrocarbons over a very long period of time.

There were three objectives in this study. First, to assess the effects of exposure to oil sand upon the composition of benthic invertebrate communities. Sites above and below the oil sands deposits were sampled qualitatively in each season over one year. Second, oil sand and rubble in a riffle in the lower part of the river were sampled quantitatively during the open-water season to compare standing stocks and the variety of benthos on each substrate. Third, since the lowermost riffle site was periodically flooded during periods of high discharge of the Athabasca River, an unflooded riffle was also sampled to examine the effects of a fluctuating current regime on the benthic fauna.

2.2.1 The Study Area

The Steepbank River is the first major tributary of the Athabasca River, entering from the east downstream of Fort McMurray, Alberta (57°02'N, 111°30'W). For most of its 116 km length (Figure 20), the Steepbank River has a gradient of 2.4 $m \cdot km^{-1}$ and

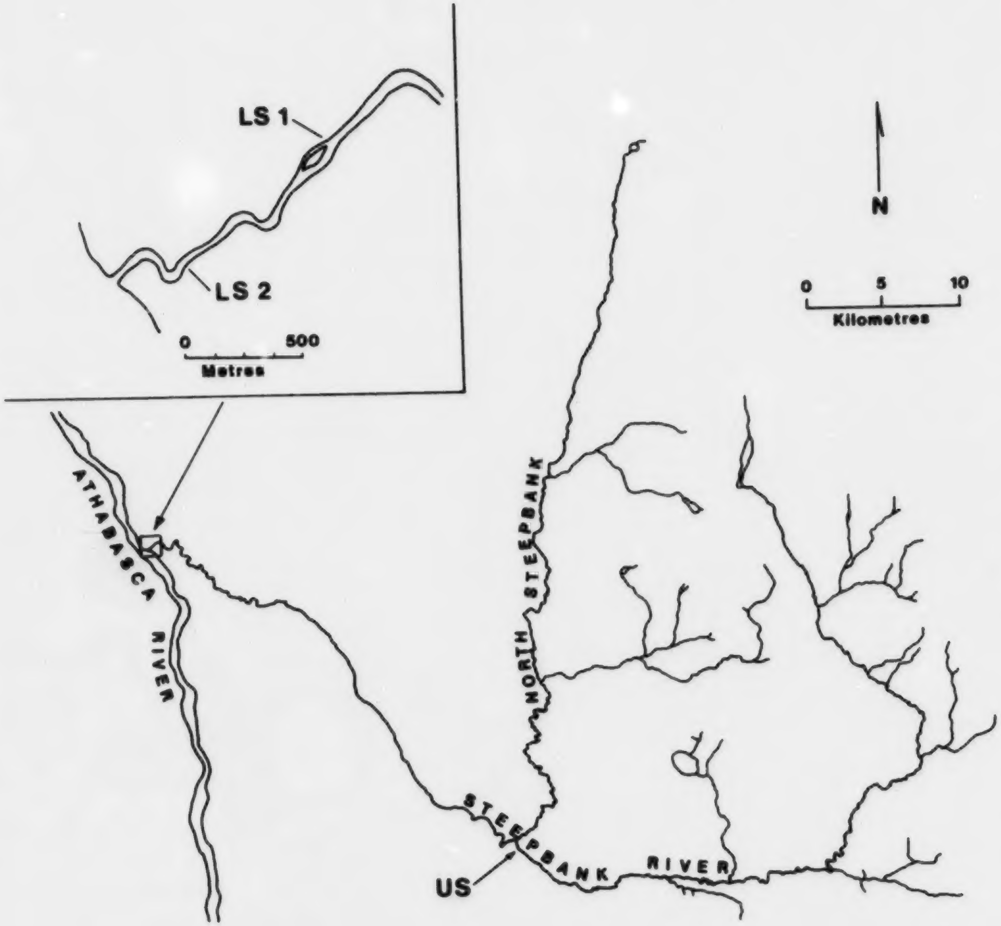

Figure 20. Locations of sites used in study of the effects of oil sand in the Steepbank River.

consists of riffles and pools with glacial gravel and boulder substrates. In its lower 20 km, the Steepbank River has a gradient of about 5.7 $m \cdot km^{-1}$ and consists of long riffles and runs with few true pools. This portion of the river cuts through the Athabasca Oil Sands and the underlying Devonian limestone so that the bed of the stream consists of a nearly continuous trough of oil sand with varying amounts of gravels and limestone rubble lying on or in it.

The mean discharge of the Athabasca River in winter is about 200 $m^3 \cdot s^{-1}$ and in summer varies from about 570 to well over 1700 $m^3 \cdot s^{-1}$. This fluctuating discharge results in changes of 2 to 4 m in the elevation of the river surface over periods as short as a few hours.

When the discharge of the Athabasca River exceeds about 1130 $m^3 \cdot s^{-1}$, the water level of the lower Steepbank River rises and the current regime is altered in the lower reaches. If the discharge of the Steepbank River is less than 7 $m^3 \cdot s^{-1}$ during such periods, current speeds are negligible as much as 500 m above the mouth and this former riffle area becomes a long pool where silts and clays are deposited. If the discharge of the Steepbank River exceeds 7 $m^3 \cdot s^{-1}$ during high water episodes, a distinct current is maintained further downstream, but the flow is much less turbulent than during low water. This alteration of riffle and pool conditions continues throughout the open-water period.

An upstream site (designated US = Site S-6), above the point where the river begins to cut through oil sand, was located 50 km above the mouth near the confluence of the North Steepbank River (Figure 20). Two riffles, 1000 and 400 m above the mouth of the Steepbank River, were chosen for the downstream sites (designated LS-1 and LS-2, respectively). LS-1 was not affected by fluctuations in the level of the Athabasca River during 1976.

2.2.2 Methods

Qualitative comparison of the fauna at the upstream (US) and downstream (LS-1) sites was based on the methods described in

Section 1.1. Rubble and oil sand as substrates, and the effects of alteration in the current regime, were studied at Sites LS-1 and LS-2 using a Surber sampler which enclosed an area of 0.09 m^2 and was equipped with a 202 μm mesh collecting bag. Oil sand substrates enclosed by the sampler were scrubbed clean with a vegetable brush. On rubble substrates, stones were removed by hand to a depth of 10 cm, or down to solid oil sand, and scrubbed. Any remaining finer sediments were agitated thoroughly to dislodge benthic organisms. When lentic conditions prevailed at Site LS-2, 0.09 m^2 quadrats were sampled using an airlift equipped with a 202 μm mesh collecting bag (Barton and Hynes in press). The sampling program is summarized in Table 6.

All samples were preserved with 10% formalin in the field and returned to the laboratory where macro-invertebrates were sorted from the associated debris under 10X magnification. All insects, except Chironomidae, were identified and enumerated at the generic or specific level. Chironomids were separated only to the level of sub-family or tribe, since most specimens had globules of tar adhering to them preventing further identification. Attempts to remove the tar with acetone and xylene were unsuccessful. The Oligochaeta and Acari were treated as individual taxa. Most of the Oligochaeta were Enchytraeidae and *Nais behningi*.

Since variances tended to be large, the data from Sites LS-1 and LS-2 were transformed as $\ln$ (number/sample + 1) (Elliot 1971). Student's t test was used to test the significance ($p < 0.05$) of differences between sites and between substrates.

2.2.3 Results

2.2.3.1 Upstream site (US) versus area of oil sand exposure (LS-1).

From the two sites, 124 taxa were collected. Of these, 17 were found only at the upstream site on two or more visits, and seven only at the downstream site (Table 7). Three of 11 stonefly species and half half of 16 caddisfly species were found only at the upstream site

Table 6. Summary of sampling at downstream sites, 1976.

DATE	SITE LS-1	SITE LS-2	CONDITIONS AT LS-2
23 June	NS[a]	4 O[b], 4 R[c]	Riffle
3 July	4 O, 4 R	2 O, 2 R	Pool
4 August	3 R	3 O, 3 R	Riffle
22 August	NS	3 O, 3 R	Pool
15 September	4 R	1 O, 3 R	Deep Riffle
4 October	4 R	3 O, 3 R	Riffle

[a] Symbol: NS = Not sampled

[b] Symbol: O = Oil sand

[c] Symbol: R = Rubble

Table 7. Taxa collected only at sites above (US) or within (LS-1) the area of eroding oil sand on two or more visits during the qualitative survey.

FAUNA[a]	SITE US	SITE LS-1
Ephemeroptera (22)	*Ephemerella tibialis*	*Ephemerella inermis*
Plectoptera (11)	*Leuctra* cf. *sara*	*Isoperla ?fusca*
	Claassenia sabulosa	
	Pteronarcella regularis	
Trichoptera (16)	*Rhyacophila*	--
	Glossosoma spp.	
	Wormaldia gabriela	
	Arctopsyche ladogenisis	
	Ceraclea spp.	
	Lepidostoma	
	Brachycentrus spp.	
	Micrasema	
Corixidae (9)	--	*Sigara bicoloripennis*
		S. conocephala
		S. solensis
Chironomidae (37)	*Microtendipes* cf. *pedellus*	*Ablabesmyia*
	Orthocladiinae A	*Paramerina*
	Heterotrissocladius cf. *marcidus*	
	Nanocladius cf. *rectinervis*	
	Synorthocladius	

[a] Numbers in parentheses indicate the total number of taxa collected.

during these regular visits. Three species of *Sigara* (Hemiptera) were found only at the downstream site. The number of taxa collected per visit was consistently greater at the upstream site (Table 8).

The percent composition of the fauna at each site showed considerable variation between visits (Table 8). While the mean percent abundance of several groups differed greatly between sites, only the Plecoptera and Trichoptera were consistently more abundant at the upstream site. Tanypodinae and Empididae (which accounted for 90% of 'other diptera') were consistently more abundant at the downstream site.

2.2.3.2 Oil sand versus rubble. During the first three months of 1976, the mean discharge of the Athabasca River was about 198 $m^3 \cdot s^{-1}$. In early April, the river rose, cresting at 1700 $m^3 \cdot s^{-1}$ on 18 April, and was clear of ice by 19 April. Discharge declined rapidly in the week following break-up and then fell slowly until mid-June.

The discharge of the Steepbank River followed a similar pattern during the first half of 1976. After a winter mean of 0.45 $m^3 \cdot s^{-1}$, the river crested at 17.7 $m^3 \cdot s^{-1}$ on 15 April, was free of ice by 20 April, and fell steadily until late May. Summer discharge is shown in Figure 21.

Flooding of the lower site due to high discharge of the Athabasca River occurred from 26 June and from 8 August to 17 September (Figure 21). Lentic conditions existed at the lower site during the first period of high water and during the second until 27 August when an exceptionally heavy rainstorm caused the discharge of the Steepbank River to increase sufficiently to reinstate lotic conditions for the rest of the open-water season.

Since the discharge of the Steepbank River was greater than 8.5 $m \cdot s^{-1}$ during April when the Athabasca River was also high, the fauna at the lower site (LS-2) was exposed to lotic conditions continuously through the winter and spring. It was assumed,

Table 8. Percent composition and number of taxa collected from sites above (US) and within (LS-1) the area of eroding oil sand.

	22 Jul 76		11 Oct 76		30 Jan 77		1 May 77		22 Jul 77	
	US	LS-1	US	LS-1	US	LS-1	US	LS-1	US	LS-1
Lower Phyla	0.2	*[a]	3.9	0.6	3.9	8.4	2.1	8.0	2.5	4.3
Oligochaeta	0.3	*	1.5	*	4.0	1.4	5.3	5.8	13.7	4.2
Ephemeroptera	29.9	19.5	20.2	34.8	13.7	5.5	18.0	41.6	37.2	44.8
Plectoptera	2.7	*	13.1	4.3	2.7	*	1.3	*	4.9	1.5
Trichoptera	22.4	*	35.0	*	34.2	*	7.5	*	5.5	3.1
Tanypodinae	0.1	3.1	3.0	5.0	0.8	0.9	1.7	4.4	0.8	5.9
Chironomini	0.5	1.8	0.9	8.9	0.9	0.2	3.0	0.8	0.4	0.5
Tanytarsini	7.2	46.1	6.3	5.1	11.7	66.5	30.9	12.7	20.0	18.5
Orthocladiinae	34.8	14.3	6.0	8.8	14.9	1.4	18.0	12.9	11.5	7.6
Other Diptera	4.7	14.4	7.2	11.5	11.0	15.0	2.5	11.7	1.6	7.0
Other Insects	0.1	*	2.1	13.9	*	*	*	0.8	0.4	0.5
Number of Taxa	46	18	58	32	47	14	48	37	47	42

[a] Symbol: * = Not sampled.

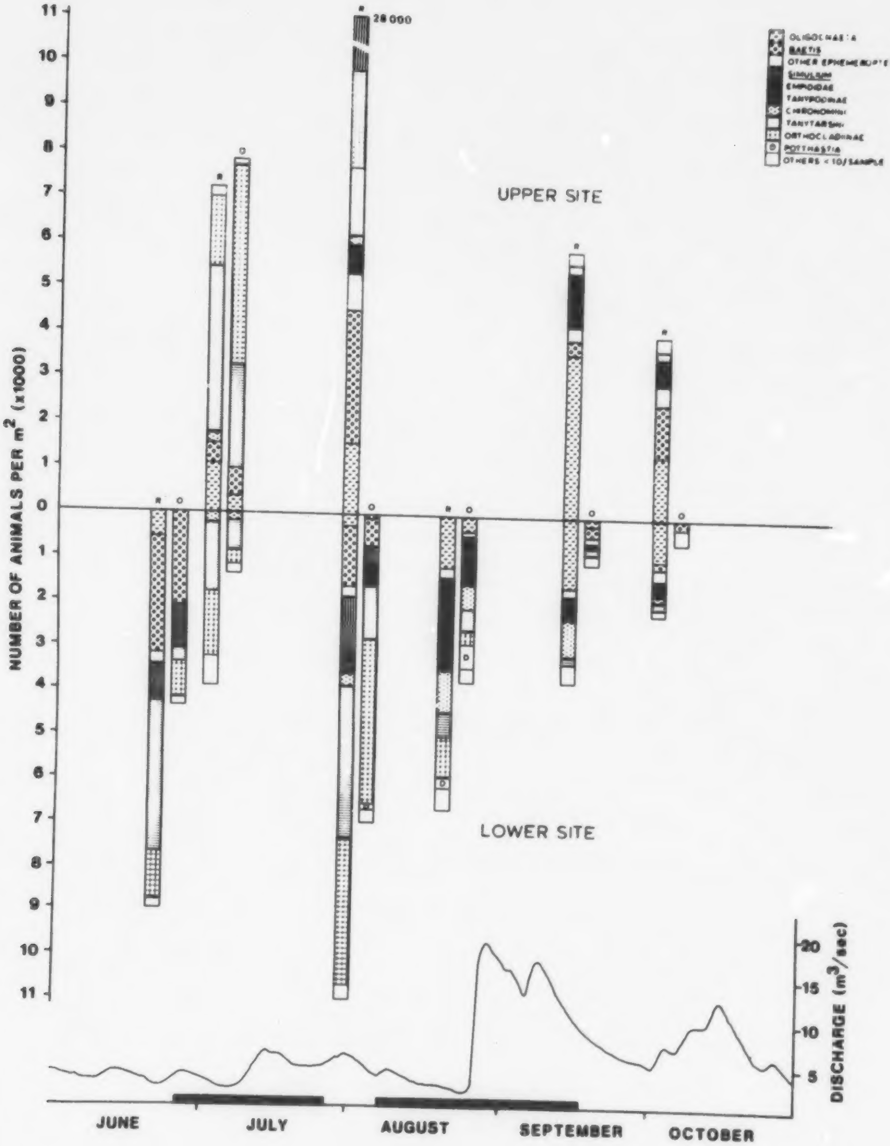

Figure 21. Mean standing stocks of invertebrates on rubble (R) and oil sand (O) at the upper (LS-1) and lower (LS-2) sites. Discharge at the lower gauging site of the Steepbank River is given at the bottom. Black horizontal bars indicate periods when the discharge of the Athabasca River exceeded 1130 $m^3 \cdot s^{-1}$.

therefore, that the fauna sampled at LS-2 in late June was similar to that of other riffles in the lower Steepbank River at that time and could be used to assess subsequent changes at both LS-1 and LS-2.

The total density of benthic invertebrates on rubble at LS-1 averaged 8300 animals$\cdot$m^{-2} in late June and early July, rose to 28 000$\cdot$m^{-2} in early August, and declined in September and October (Figure 21). The high densities recorded in early August were associated with the appearance of second generations of *Baetis* and *Simulium tuberosum*-complex. The autumn decline in standing stocks was probably due to the completion of the emergence of many of the insect species and the scouring effects of the spate in late August and early September. The number of non-chironomid taxa per sample showed a similar peak in August, but the late summer values were slightly higher than those for June and July (Table 9).

Flooding and the imposition of lentic conditions at LS-2 was accompanied by a 50% reduction in the total benthic standing stock in both July and August (Figure 21). This was largely due to the elimination of such rheophilic forms as *Baetis*, *Simulium*, and *Rheotanytansus*. Invertebrate density decreased in a pattern similar to that observed at the upper site following the spate of early September.

Total invertebrate standing stocks were significantly smaller at LS-2 on dates when both sites were sampled (Table 10). Among the major groups of organisms, the mean densities of Oligochaeta, *Baetis*, and *Simulium*, were significantly greater at LS-1, and Chironomini were significantly more abundant at LS-2 (Table 11). There was no difference between sites in the mean number of taxa per sample (Table 9) or in the total number of taxa collected on any given date.

Both the variety and density of invertebrates on oil sand were significantly less than on rubble substrates (Tables 9 and 10). This did not appear to be due to a toxic effect from the oil sand since all but a few rare taxa were collected from both substrates. Burrowing or negatively phototropic forms, such as the Oligochaeta,

Table 9. Mean numbers of taxa per sample (excluding Chironomidae) collected at the lower riffle sites.

Site	Substrate	23 Jun	3 Jul	4 Aug	22 Aug	15 Sep	4 Oct
LS-1	Rubble	-	9.5	18.7	-	12.0	12.2
LS-2	Rubble	10.5	7.5	17.0	15.7	13.0	11.0
LS-2	Oil sand	4.2	4.5	12.0	11.3	-	6.3

Table 10. Mean numbers of invertebrates per sample (0.09 m^2) and results of t-comparisons. between substrates.

Site	Substrate	23 Jun	3 Jul	4 Aug	22 Aug	15 Sep	4 Oct	t	df
LS-1	Rubble	NS[c]	654.2	2527.3	NS	546.8	376.8		
								2.40[a]	1.24
LS-2	Rubble	805.2	344.5	974.0	599.7	335.0	194.3		
								4.38[b]	1.28
LS-2	Oil sand	392.0	121.5	626.7	336.7	NS	50.0		

[a] Probability $p < 0.05$

[b] Probability $p < 0.01$

[c] Symbol: NS - not sampled.

Table 11. Mean numbers of organisms in major groups and results of t-comparisons between the lower riffle sites (df = 26) and between substrates (df = 35) based on all samples collected.

	Sites					
	LS-1	LS-2	t	Rubble	Oil Sand	t
Oligochaeta	170.8	57.4	3.50^b	60.7	15.2	4.52^b
Baetis	97.6	34.4	3.27^b	86.6	63.0	0.50
Other Ephemeroptera	26.8	15.7	0.26	15.1	3.7	4.85^b
Plectoptera	4.7	2.4	0.91	3.8	0.5	5.32^b
Trichoptera	3.1	1.2	0.80	1.5	0.9	0.53
Simulium	340.4	18.5	1.79^a	31.3	25.0	0.40
Empididae	41.6	21.3	1.18	30.4	17.3	1.25
Tanypodinae	11.3	18.5	0.69	21.7	7.8	2.02^a
Chironomini	10.4	30.1	1.73^a	26.4	11.5	2.85^a
Tanytarsini	121.8	107.8	0.56	217.9	81.6	2.67^a
Orthocladiinae	124.2	94.4	0.52	129.7	161.8	0.39
Diamesinae	4.2	2.7	1.35	5.3	10.3	0.48

^{a}P <0.05

^{b}P <0.01

Plecoptera, the mayflies *Ephemerella*, *Heptagenia*, and *Rhithrogena*, and all Chironomidae except Orthocladiinae, were significantly less abundant on oil sand, while surface dwelling forms such as *Baetis* and *Simulium* were not (Table 11). Since Tanytarsini arrange their tubes to face into the current, whether on the top, sides, or bottom of stones, the lower density on oil sand was probably due to the smaller total surface area rather than lack of a preferred microhabitat.

During the more extensive sampling at the downstream sites (LS-1 and LS-2), small populations of several taxa were found which had been recorded only at the upstream site during the seasonal, qualitative study (Table 7). These included *Leuctra* cf. *sara*, *Glossosoma*, *Arctopsyche*, *Ceraclea*, *Lepidostoma*, *Micrasema*, *Microtendipes pedellus*, *Nanocladius*, and *Synorthocladius*.

2.2.4 Discussion

The effects of oil on freshwater benthos have been reported to be quite variable, depending on such factors as the type of oil, the season of exposure, and the nature of the receiving water, lentic or lotic (McCauley 1966; Parker et al. 1976). Several general trends are apparent, however. Lighter oils appear to be highly toxic to most benthic invertebrates but disappear rapidly through evaporation (Snow and Rosenberg 1975a). Certain Trichoptera, Chironomidae, Plecoptera, and Ephemeroptera show a long-term susceptibility to heavier oil fractions, while other Diptera, especially Orthocladiinae, are very tolerant and may even increase in abundance on oiled substrates (Bengtsson and Berggren 1972; Anon. 1973; Snow and Rosenberg 1975b; Rosenberg and Wiens 1976; Barton and Wallace in prep.). Oil tends to accumulate in lake sediments where its effects persist over long periods of time (Bengtsson and Berggren 1972; Snow and Rosenberg 1975b; Hare 1976), but is rapidly flushed from rivers (Chen et al. 1976) where the fauna may recover in a year (Snow et al. 1975; Rosenberg and Wiens 1976) or less, at least partially (Anon. 1973).

Bitumen, the hydrocarbon component of oil sand, is a heavy, tar-like substance which has been suggested to be the result of weathering and microbial decomposition of conventional crude oils (Rubenstein et al. in prep.). The relatively small proportion of soluble light oils in the bitumen and their rapid evaporation from turbulent river water probably accounts for the lack of a gross toxic effect from oil sand in the Steepbank River. However, the present results show that the variety of benthic organisms inhabiting the portion of the river which cuts through the oil sands deposit is smaller than in upstream areas; qualitative sampling consistently yielded more taxa at the upstream site (US). Among the groups which appear to be most affected by exposure to oil sand, the Plecoptera and Trichoptera stand out both in terms of a reduction in the variety of forms and in their numerical contribution to the entire fauna.

The physical behaviour of eroding oil sand is probably of primary importance in this regard, though there is evidence that oil is toxic to some caddisflies and stoneflies (Bugbee and Walter 1973; Snow and Rosenberg 1975a). Where streams erode banks of oil sand, some of the material breaks off as large chunks, but it is also carried downstream as minute, tar-covered particles. The relative absence of Trichoptera, especially the net-spinning Hydropsychidae and Philopotamidae, in the lower Steepbank River is probably related, at least in part, to these suspended particles of oil sand. This material may interfere with the operation of the feeding nets or may be toxic if ingested by the larvae while cleaning the net. The different feeding mechanism of *Simulium*, the other dominant filter-feeder in the Steepbank River, may be less susceptible to fouling by particles of tar and allow them to do well in the absence of competition from caddisfly larvae (Davies 1950).

Large chunks of oil sand behave somewhat differently. Where there is no obstacle to hinder their movement, isolated pieces are moved downstream by the current and are worn into shapes identical to fluvial gravel and pebbles. If these come to rest on the stream bed, they are colonized by the normal benthic fauna. Large quantities of oil sand entering a stream, as from a major bank slippage, tend to fuse together and very slowly flow downstream. Such fusion and downstream movement leads to the formation of the impermeable tarmac-like layers of oil sand which form much of the substrate in the lower Steepbank River.

As a substratum for benthic invertebrates, this layer of fused oil sand is analogous to bedrock where the density and diversity of the fauna is limited by the effective surface area available for colonization, and the lack of sheltered surfaces and a hyporheic refuge (Williams and Hynes 1974). The mean count of 3400 animals$\cdot$m^{-2} (1400$\cdot$m^{-2} if chironomids are excluded) is similar to the standing stocks on bedrock substrates reported by Pennak and van Gerpen (1947), Armitage (1961), and Barton and Hynes (1978), despite differences in sampling techniques. Such comparison further emphasizes the lack of an overall toxic effect from exposure to oil sands. Certainly these results are more similar to each other than to the mean standing stocks of 250 000$\cdot$m^{-2} in streams with porous substrates studied by Hynes et al. (1976).

Scott and Rushforth (1959) hypothesized that large stones lying on the stream bed allow the development of larger benthic standing stocks since the large stones provide areas of reduced current, protection from vertebrate predators, accumulations of organic debris, and stability during spates. This is a specific example of the more general theory that habitat complexity and faunal diversity are directly proportional (MacArthur and MacArthur 1961). The present results support this theory: the less structurally complex habitat afforded by oil sand supported fewer animals of fewer kinds than did the physically varied rubble substrates.

It was expected that the more stable current regime at Site LS-1 would support a greater diversity of organisms than was found at Site LS-2, which was subject to wide fluctuations in this environmental parameter (Klopfer 1959; de March 1976). This was not the case over the summer as a whole, although the number of species per sample is perhaps not the best measure of diversity (Wilhm 1967; de March 1976). Since the Chironomidae were seldom identifiable, calculation of more elaborate diversity indices was not considered appropriate. When considering the effects of alternating lotic and lentic conditions, the lack of an overall difference in the variety of animals between Sites LS-1 and LS-2 suggests that, while several strongly rheophilic forms such as *Baetis*, *Simulium*, and *Rheotanytarsus* were largely eliminated by reductions in current speed, many of the species inhabiting the lower Steepbank River are capable of surviving severe changes in current velocity, at least for periods less than an entire season.

In summary, the variety and percentage composition of stream benthos were altered in the portion of the lower Steepbank River which cuts through the Athabasca Oil Sands deposit. The relative importance and variety of Trichoptera and Plecoptera were less in this downstream section of the river. These differences in the benthic communities were probably largely caused by physical alteration of the substrate and the presence of particles of oil sand in suspension. The standing stocks of benthic invertebrates on oil sand substrates were about half as large as those on rubble and contained significantly fewer burrowing or negatively phototropic forms. Flooding of riffle habitat resulted in a 50% reduction in benthic standing stocks, but the community recovered rapidly upon the resumption of normal current.

3. BASELINE SURVEY OF THE MACROBENTHOS OF THE ATHABASCA RIVER

3.1 INTRODUCTION

The Athabasca River has been the focus of human activity in northeastern Alberta since Peter Pond first explored the area in 1778. Within the past few decades, the nature of human activity along the lower reaches of the river has increasingly shifted from trading and transportation to industrial development of the Athabasca Oil Sands. Since the river is the major source of fresh water and is the ultimate recipient of all surface runoff from the oil sands area and is vital to the ecology of the Peace-Athabasca delta, it is essential to gain an understanding of its biological properties. Previous studies have concentrated on fish resources, bacteriological degradation of hydrocarbons, and benthos, especially with respect to attempts to control nuisance outbreaks of blackflies (Bond in prep.; Geesey and Costerton in press; McCart et al. 1977; Flannagan et al. in prep.).

Despite these efforts, there remains little published information concerning the abundance and species composition of invertebrates directly inhabiting the bed of the Athabasca River, or indeed any large river in North America. As a first step toward filling this gap in the understanding of aquatic ecosystems, a program of direct sampling of benthos at six sites in the vicinity of Fort MacKay, Alberta, was undertaken on a monthly basis from June through October 1977. This section describes the results of that study, with special emphasis on the effects of substrate on the development of benthic communities and techniques for sampling those communities.

3.1.1 Methods

Five sampling stations were established along the west bank of the river in mid-May 1977 (Figure 22). Each was marked with a styrofoam mooring buoy anchored by a 25 L pail of concrete with a 1.3 m length of steel fence post embedded transversely through it. Three cylindrical stainless steel baskets (Anderson and Mason 1968),

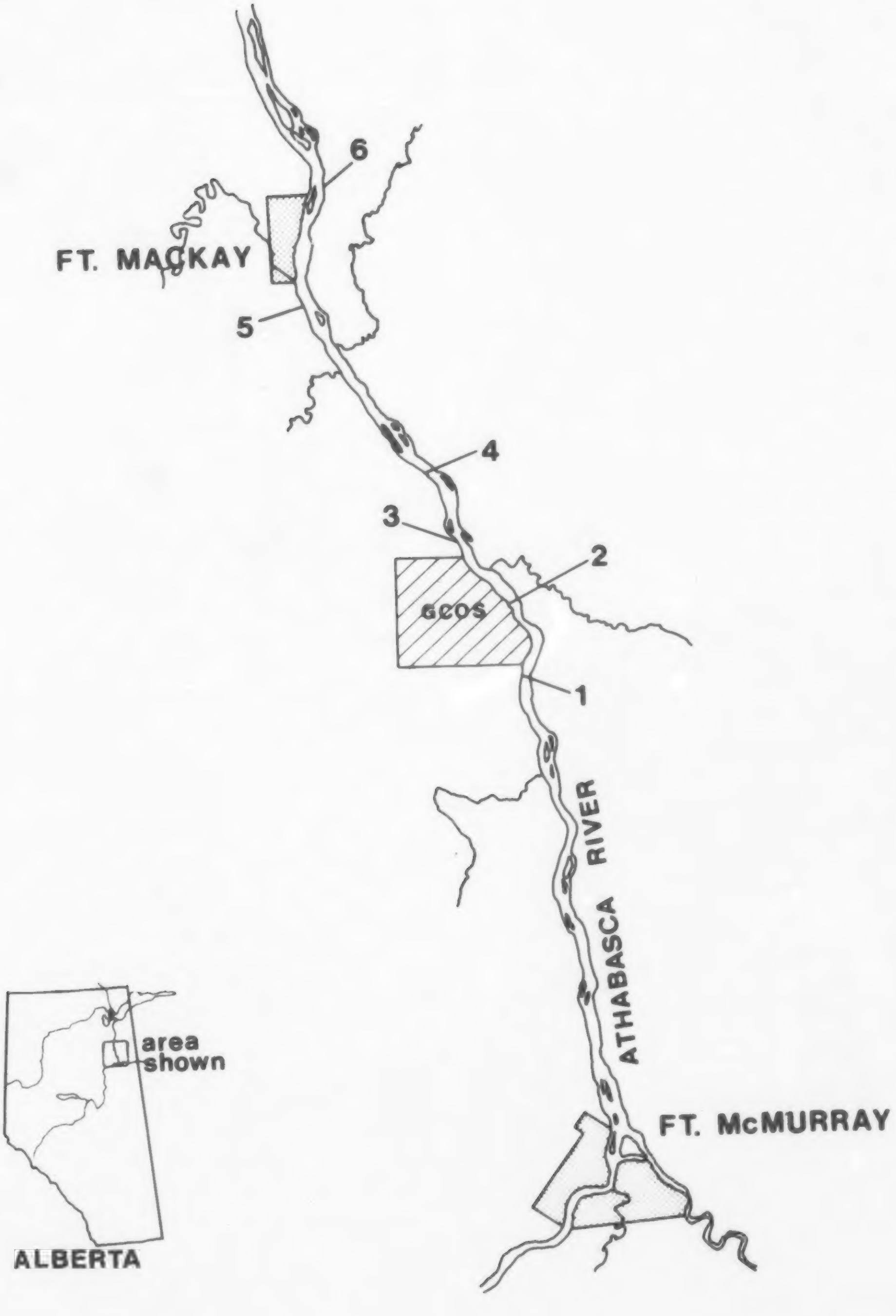

Figure 22. Locations of study sites on the Athabasca River.

each containing seven or eight cobbles 8 to 10 cm in diameter, collected from a nearby deposit of glacial gravel, were attached to a line, dropped to the bottom of the river, and tied to each buoy. A sixth site was chosen on an outcrop of Devonian limestone but neither buoy nor baskets could be stationed here since it was within the navigation channel.

Four weeks later, on 21 June, the buoys at Site 4 and 5 had disappeared and the baskets at Site 2 had been buried by sand and could not be raised. The baskets at Sites 1 and 3 were pulled slowly to the surface and enclosed in a 200 μm mesh net before being lifted into the boat. The upstream sides of the baskets were covered with varying amounts of organic debris (sticks, leaves, grass, etc.) and close inspection revealed that very few, if any, organisms were living on the stones although the debris contained large numbers of benthic animals. Since it was impossible to separate the animals associated with the debris from those associated with the rocks or to estimate how long the debris had been on the baskets, the use of artificial substrates was abandoned. In late August, it was discovered that the buoys at Sites 4 and 5 had been dragged beneath the surface by the baskets which had been rolled downstream. Even after three months in the river, essentially no animals appeared to be living on the rocks, but the debris on the outside of the baskets and the mooring lines contained a rich fauna.

A 15 cm^2 Ekman grab and an airlift were used to take three samples from mud or coarser sediments at each site at approximately four-week intervals from June through October. The airlift was constructed of 5 cm i.d. aluminum pipe in sections of various lengths which could be screwed together as needed, according to the depth at the site. The nozzle (Barton and Hynes in press) was built into one section and the top section had two 90° bends with a rim around the final aperture to hold the drawstring of a 202 μm mesh collecting bag. A SCUBA tank was used as an air supply. The airlift was operated over the stern while the boat was tied at the bow to the buoy marking the sampling site. The pipe was held vertically in the water column with the aid of two ropes fastened near the nozzle. These passed forward through pulleys bolted to the ends of a boom across the bow and were tied off on cleats on the middle seat of the boat. Two people were required for the operation of the airlift: one to control the flow of air from the valve on the SCUBA tank and to tend the stabilizing lines, the other to move the pipe vertically and horizontally along the bottom of the river. The area sampled was estimated from the volume of sand collected in the bag, assuming an effective sample thickness of 5 cm.

Samples collected in this manner were emptied into a bucket of river water, agitated thoroughly, and the water and suspended organisms decanted through a 180 μm mesh sieve. The process was repeated seven times or until no new organisms appeared on the sieve after three additional successive decants. Samples of mud and muddy sand taken with the Ekman grab were transferred to 202 μm mesh bags and washed to remove the sediments. The residue on the sieve or in the bag was preserved immediately in 10% formalin.

Debris was collected from the anchor line of the mooring buoys and organisms were hand-picked into 70% ethanol. A portion of the debris was washed in river water. The washings were concentrated using a 180 μm mesh sieve and preserved in 10% formalin.

Organisms living at depths of 2.5 to 3.0 m on bedrock at Site 6 were collected with the airlift while the boat drifted with the current. It was impossible to estimate the area sampled due to the irregularity of the surface of the rock. Two samples, each representing about two minutes of airflow, were taken each month. Additional material was collected with a 500 μm net along sandy and rocky shores at least monthly throughout the open-water season. These animals were used to supplement life history analyses.

Organisms were sorted from the associated debris in the samples under 10X magnification in the laboratory. All animals, except Nematoda, Cladocera, Ostracoda, Copepoda, Acari, and Enchytraeidae, were enumerated at the generic or specific level. Community diversity ($\bar{d}$) was calculated using a computer program based on that given by Wilhm (1970).

Records of turbidity and daily discharge of the Athabasca River below Fort McMurray during 1978 were obtained from the Inland Waters Directorate, Water Survey of Canada.

3.1.2 Results

The discharge of the Athabasca River averaged about 210 $m^3 \cdot s^{-1}$ during the first three months of 1977, and increased to 1217 $m^3 \cdot s^{-1}$ before the ice broke on 29 April. Daily discharge from May through October is shown in Figure 23. The temperature rose from near 0°C under ice-cover to 12° by late May and over 18° during June, July, and August, then dropped steadily from September until ice covered the river on 12 November. Turbidity increased with discharge, ranging from 1 to 8 Jackson units under ice-cover to over 100 during peak flows in June and July.

Fluctuating discharge effected changes in the nature of the sediments at Sites 2, 3, and 4 (Table 12). Erosion or deposition of sediment at each of these sites appeared to be related to a combination of discharge, topography of the river bed, and the nature of the sediments in the area.

The composition of the benthic fauna varied on each of the principle types of substrate. A total of 114 taxa were collected during this study, of which 31 appeared in over 65% of all the samples from any given substrate (Table 13). (A complete list of invertebrates is given in Appendix 6.1.) Orthocladiinae B was the most frequently collected species, occurring on all substrates except debris, and dominated the fauna on coarse sand through June and July. The proportion of rare taxa tended to decrease with increasing stability of the substrate (Table 13). Changes in the specific and percentage composition of the fauna at each site, as indicated by low values of CC and PSC, tended to be greatest during the first three months of the study (Table 14) as a result of emergence of adult insects (especially 'Orthocladiinae B') and erosion or deposition of sediments.

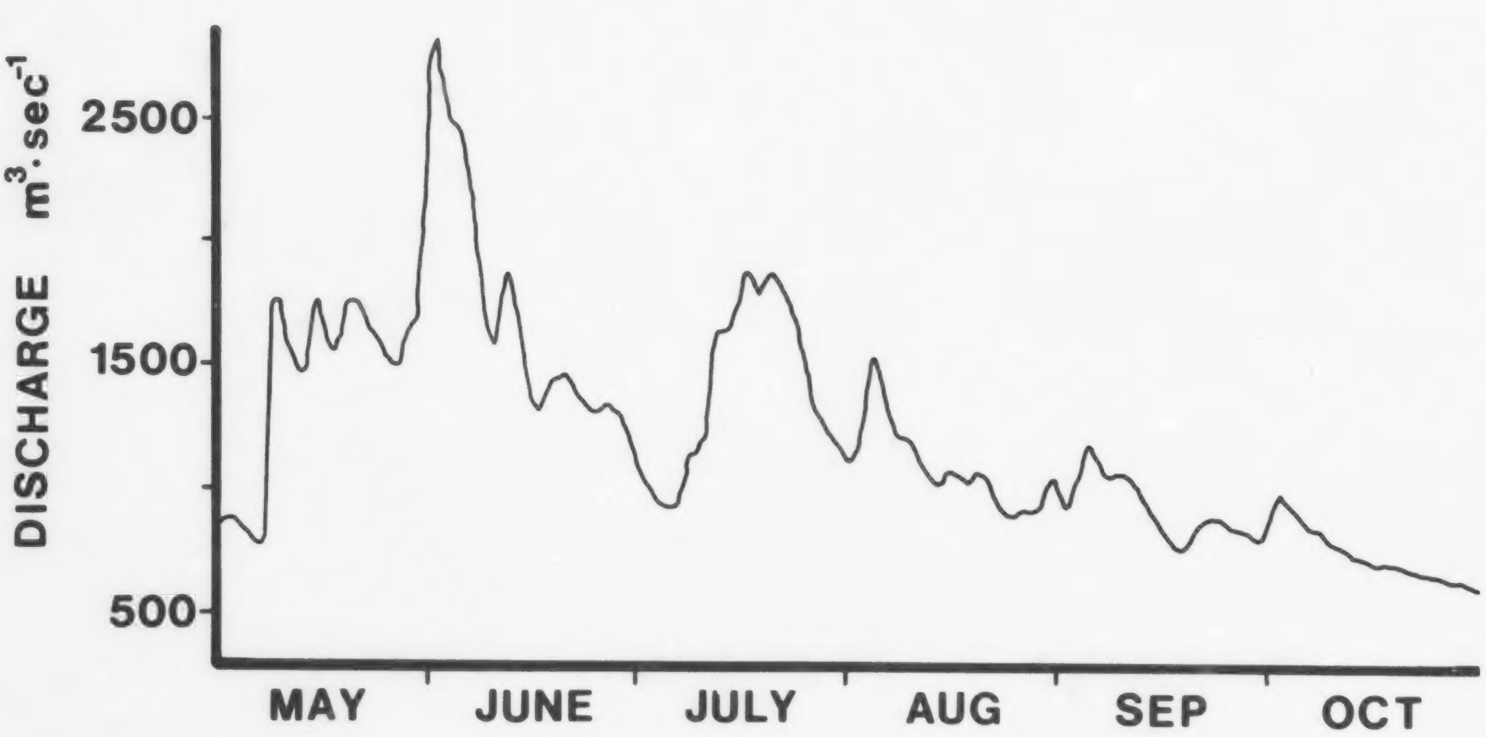

Figure 23. Discharge of the Athabasca River, May to October 1977.

Table 12. Major substrate types at Sites 1 to 5.

	Sampling Site				
Month	1	2	3	4	5
June	Coarse Sand	Muddy Sand	Muddy Sand	Fine Sand	Very Coarse Sand
July	Coarse Sand	Muddy Sand	Coarse Sand	Medium Sand	Very Coarse Sand
Aug	Coarse Sand	Muddy Sand	Muddy Sand	Medium Sand	Very Coarse Sand
Sept	Coarse Sand	Mud	Muddy Sand	Gravel	Gravel/Coarse Sand
Oct	Coarse Sand	Mud	Muddy Sand	Gravel	Gravel/Coarse Sand

SUBSTRATE TYPES

Substrates	Particle Size
Gravel	2 - 15 mm
Very Coarse Sand	1 - 2 mm
Coarse Sand	0.5 - 1 mm
Medium Sand	0.25 - 0.5 mm
Fine Sand	0.01 - 0.25 mm
Mud	<0.01 mm

Table 13. Taxa occurring in at least 65% of all samples from a single substrate.

Taxa	Mud	Sand	Coarse Sand	Gravel	Bedrock	Debris
Nematoda				+	+	
Tubificidae w/o hair setae	+				+	
Acari					+	
Ametropus neavei				+		
Baetis spp.					+	+
Ephemerella inermis					+	+
Heptagenia				+	+	+
Ophiogomphus colubrinus					+	
Oenopteryx fosketti				+		
Pteronarcys dorsata						+
Isogenus frontalis colubrinus					+	+
Isoperla longiseta				+	+	+
Hastaperla brevis				+		
Acroneuria abnormis					+	
Cheumatopsyche					+	+
Hydropsyche					+	+
Oecetis avara					+	
Simulium arcticum		+			+	
Empididae					+	
Thienemannimyia					+	

continued...

Table 13. Concluded.

Taxa	Mud	Sand	Coarse Sand	Gravel	Bedrock	Debris
Chernovskiia orbicus				+		
Cyphomella cf. *gibbera*	+					
Paracladopelma	+					
Polypedilum breviantennatum-gp.	+	+		+		
Robackia claviger		+		+		
R. demeijerei			+		+	
Cladotanytarsus					+	
Rheotanytarsus						+
Eukiefferiella spp.						+
Nanocladius					+	
Orthocladiinae B		+	+	+	+	
No. of frequently collected taxa	4	4	2	10	21	9
Commonly collected	12	2	9	17	20	6
Rare	36	15	45	20	39	41
Number of samples	28	6	33	9	6	24

Table 14. Percent Similarity of Community and (Coefficient of Community) between sampling visits.

Site	Month								
	June		July		Aug		Sept		Oct
1		58.8 (37.5)		20.4 (20.0)		57.7 (29.2)		59.2 (38.7)	
2		45.7 (29.6)		33.0 (26.3)		30.2 (32.4)		91.7 (50.0)	
3		23.6 (31.6)		5.2 (17.2)		29.2 (41.4)		41.2 (41.4)	
4		85.7 (22.7)		58.9 (20.8)		23.6 (23.5)		60.1 (51.2)	
5		61.2 (12.8)		26.7 (17.6)		62.4 (28.0)		61.2 (38.7)	
6		34.6 (44.2)		30.7 (44.0)		64.3 (50.0)		61.9 (52.0)	

Estimates of the density of the fauna at Sites 1 to 5 are summarized in Figures 24 to 26. Orthocladiinae B and *Robackia* spp. dominated the fauna of coarse sand in June and July, the former being largely replaced by *Paratendipes, Isoperla,* and *Hastaperla* in September and October, especially on the coarser grained sediments at Sites 4 and 5. *Robackia demeijerei* tended to be more abundant on coarse sand, while *R. claviger* showed a preference for gravel and fine sand.

The fauna of muddy sand at Sites 2 and 3 was dominated by oligochaetes (mostly immature Tubificidae) and larvae of *Harnischia*-gp. and *Polypedilum breviantennatum*-gp. The proportion of these chironomids increased as fine sediment continued to be deposited in the later part of the study.

Dominant invertebrates on bedrock (Table 15) included the mayflies *Baetis* spp., *Ephemerella inermis, Heptagenia,* and *Rhithrogena;* the stonefly *Isoperla longiseta;* the caddisfly *Cheumatopsyche;* larval Empididae; and the chironomids *Cladotanytarsus, Rheotanytarsus,* and *Nanocladius.* The species composition of the fauna was more consistent than on less stable substrates (higher CC values, Table 14), but changes in percent composition followed the same general pattern observed at Sites 1 to 5.

The amounts of debris caught on the anchor lines at each site in each month were too inconsistent to permit comparisons between stations. During periods of high discharge, large amounts of debris were caught on all lines. During low water, the amount of material was somewhat proportional to the strength of the current at the site, but since large sticks tended to act as nuclei for the accumulation of more debris, the relationship was not a simple one. Therefore, all data from debris collected on each date has been combined (Table 16). Most of the animals found in these accumulations of sticks, grass, leaves, and roots were Plecoptera (*Isoperla longiseta, Isogenus frontalis colubrinus, Pteronarcys dorsata*), Ephemeroptera (*Baetis, Ephemerella inermis, Heptagenia*), and in July, *Simulium arcticum.* Three species of *Eukiefferiella* composed most

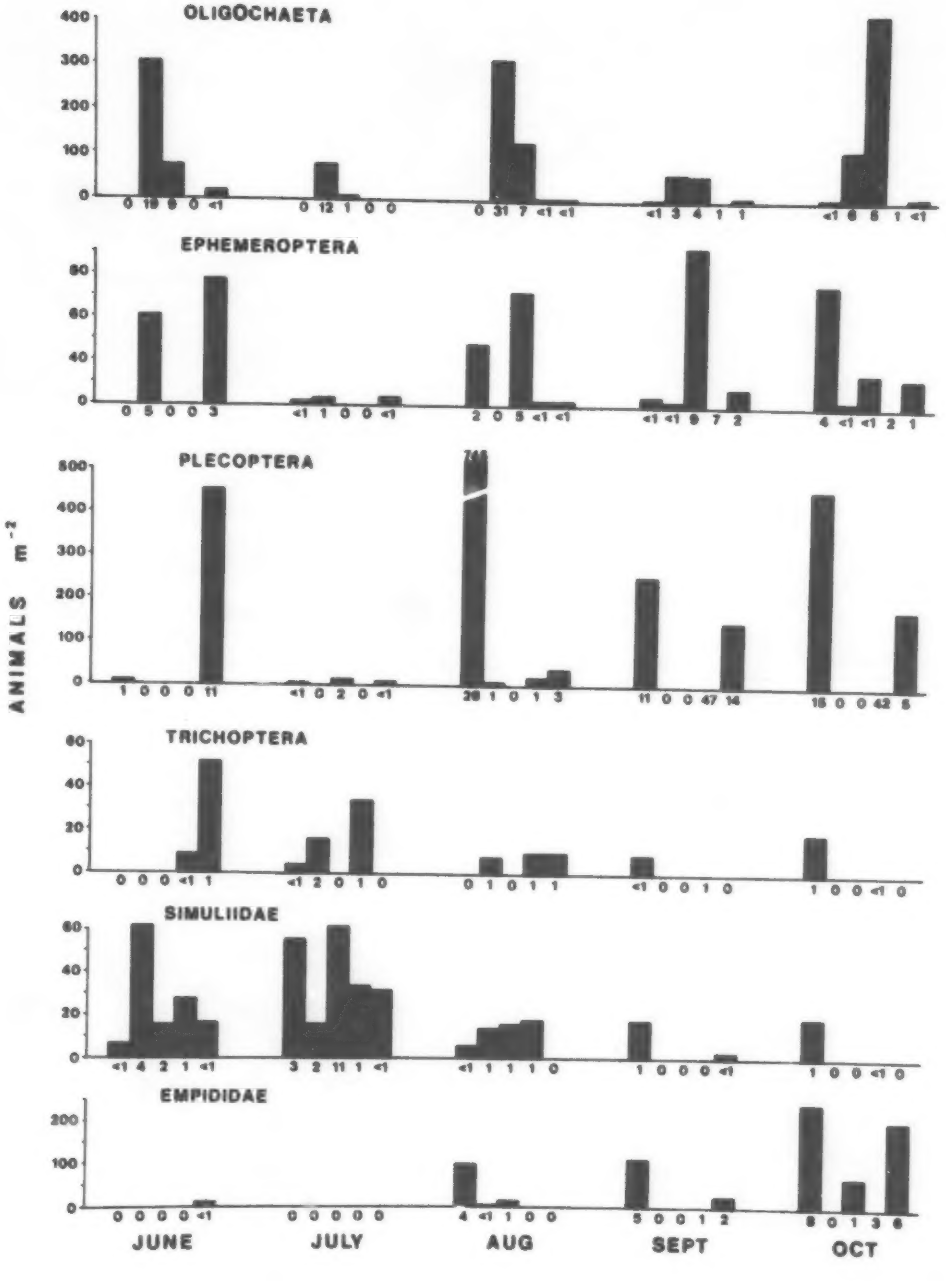

Figure 24. Mean standing stocks of invertebrate groups at Site Sites 1 to 5 (left to right, each month). Numbers below each column indicate the percentage of the total fauna in each group.

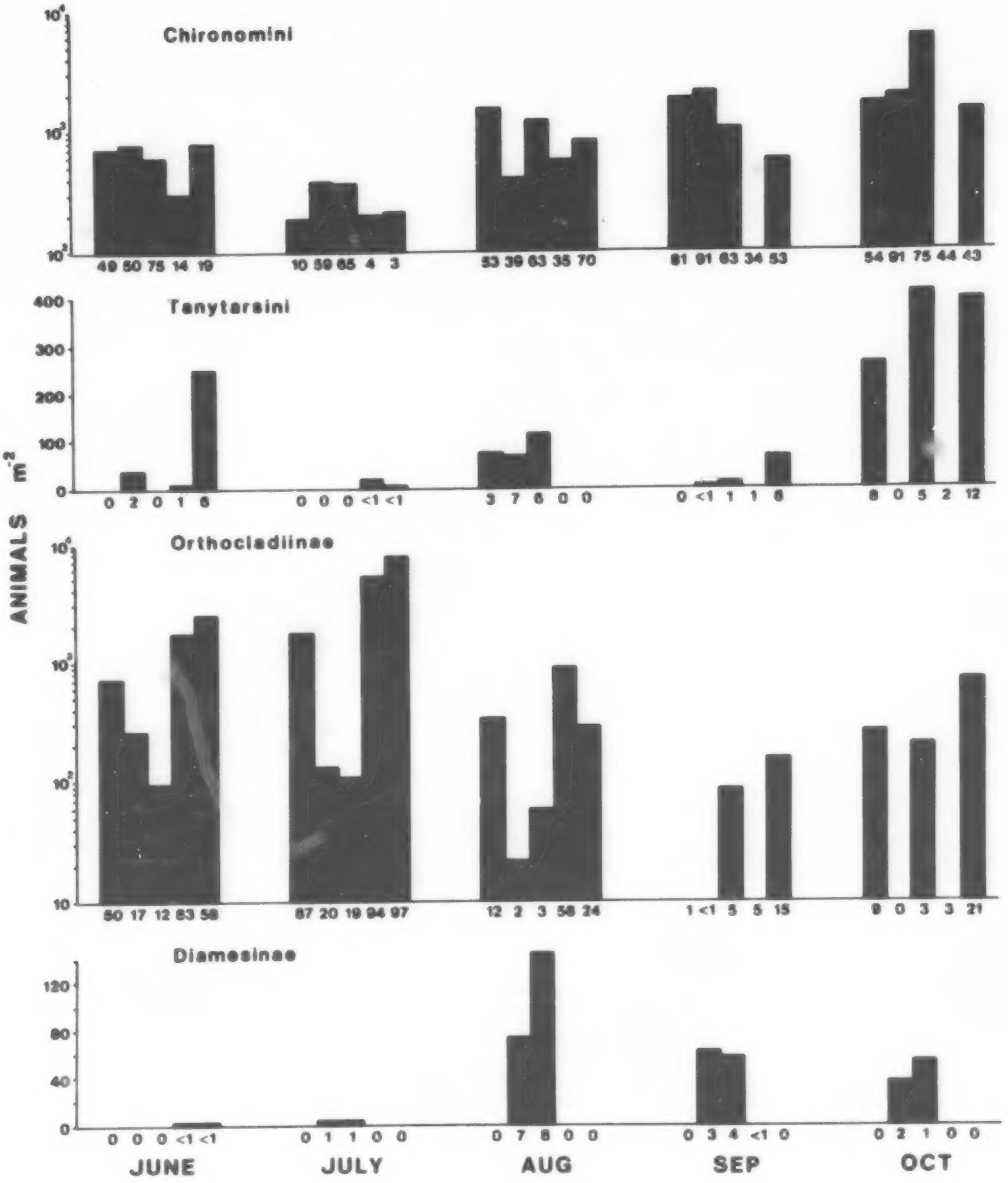

Figure 25. Mean standing stocks of tribes and subfamilies of Chironomidae at Sites 1 to 5 (left to right, each month). Numbers below each column indicate the percentage of the total fauna in each group.

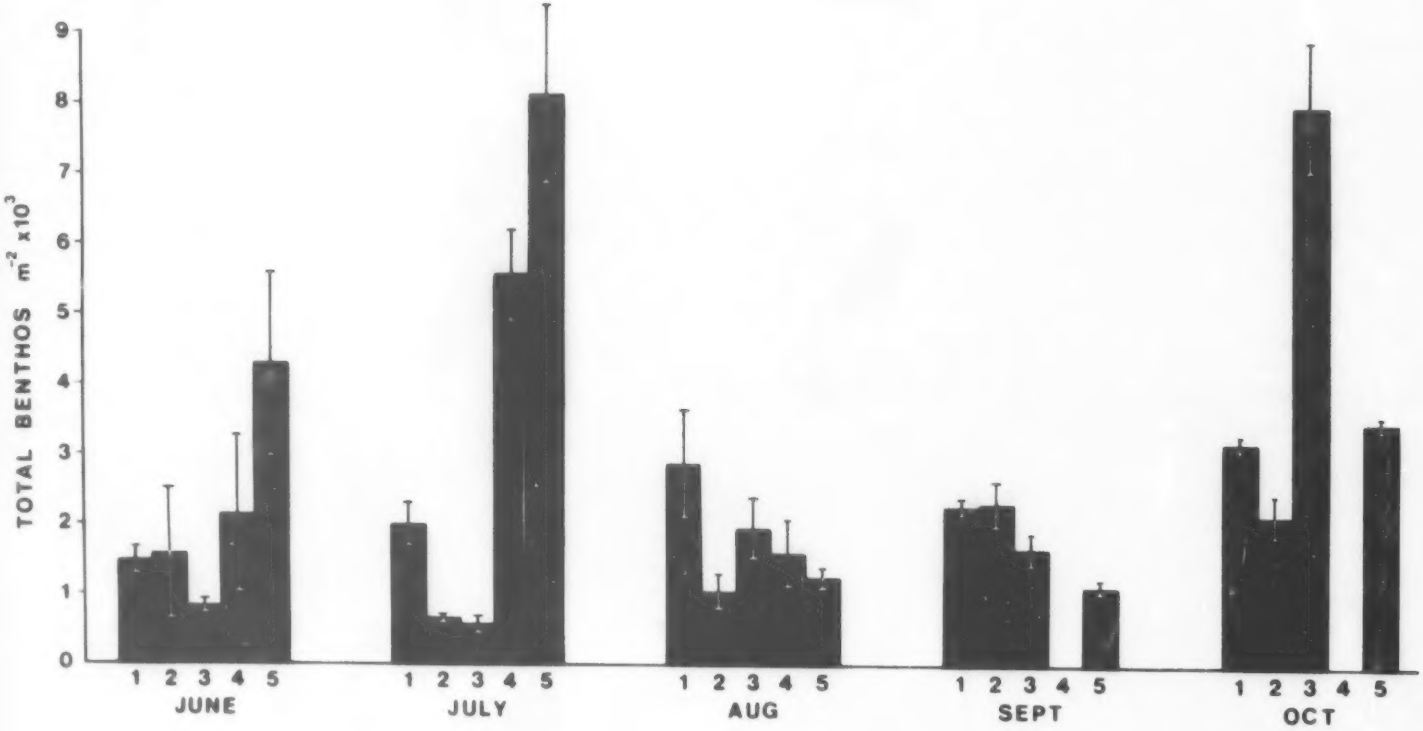

Figure 26. Mean density ± S.E. of total benthos at Sites 1 to 5. Samples from Site 4 in September and October could not be quantified.

Table 15. Number of taxa and percent composition of the fauna collected from bedrock.

Taxa	Month					
	May	June	July	Aug	Sept	Oct
Lower Phyla	1	8	3	3	5	1
Oligochaeta	2	24	2	2	5	2
Ephemeroptera	19	19	8	16	6	15
Plecoptera	30	4	11	26	11	24
Trichoptera	2	4	8	8	11	5
Tanypodinae	5	3	1	2	2	6
Chironomini	2	4	1	<1	1	5
Tanytarsini	14	12	3	11	20	23
Orthocladiinae	7	19	56	7	5	3
Other Diptera	12	2	6	21	30	16
Other Insects	2	1	1	2	2	<1
Number of taxa	54	35	40	32	42	34

Table 16. Number of taxa and percent composition of the fauna collected from debris.

Taxa	Month June	July	Aug	Sept	Oct
Oligochaeta	<1	<1	3	1	-
Ephemeroptera	30	19	9	12	11
Plecoptera	47	20	30	62	72
Trichoptera	6	4	40	15	14
Simulium	7	50	1	2	4
Tanypodinae	<1	2	1	<1	
Chironominae	1	1	<1	1	
Tanytarsini	<1	<1	-	<1	-
Orthocladiinae	7	4	14	<1	-
Other Insects	1	<1	1	1	
Number of taxa	25	29	37	33	13

of the Chironomidae. Many drifting animals probably colonized the accumulated debris but animals were also found clinging to sticks and logs floating freely downstream during high water.

Diversity ($\overline{d}$) was consistently high on bedrock (Table 17). On coarse sand, values were low but increased in autumn as discharge declined. The low values of $\overline{d}$ for mud in September and October reflected the dominance of *Polypedilum breviantennatum*-gp. and *Paracladopelma*. Muddy sand supported moderately diverse communities throughout the summer.

3.1.3 Discussion

Perhaps the most striking aspect of the results of this study is the dynamic nature of the benthic environment of the lower Athabasca River. Both the texture of the sediments and the number and variety of organisms found at Sites 1 to 5 underwent at least subtle, and in most cases substantial, changes between sampling visits. These changes appeared to be directly linked to variations in the direction and magnitude of currents at each site as discharge fluctuated through the summer, and to the life histories of the invertebrate species.

In his review of the hydrobiology of Russian rivers, Shadin (1956) emphasized the importance of both current and the nature of the sediments on the development of characteristic assemblages of benthic organisms. He described distinctive communities inhabiting substrates of rock, sand, silt, and clay in large rivers, including the Volga, the middle reaches of which appear to be physically similar to the Athabasca River in the study area (Behning 1928). With the exception of molluscs and amphipods, which were important members of most biocenoses in the Volga, the general composition of the fauna of these various substrates was very similar to that found in the Athabasca, in some cases even at the specific level.

The sand-dwelling benthos of the Volga River was dominated by Enchytraeidae, *Tubifex newaensis*, chironomid larvae of the *Harnischia*-gp. and *Polypedilum breviantennatum*, nematodes, and other

Table 17. Mean diversity ($\overline{d}$) at Athabasca River, Sites 1 to 6.

Site	Month					
	May	June	July	Aug	Sept	Oct
1	NS[a]	1.085	0.727	1.987	1.153	2.893
2	NS	2.965	2.991	3.247	1.294	1.324
3	NS	2.347	2.181	2.820	2.925	2.464
4	NS	1.038	0.513	2.245	2.888	3.217
5	NS	2.225	0.257	1.637	2.677	3.059
6	3.653	4.099	3.006	3.854	3.844	3.841

[a] Symbol: NS = Not sampled.

oligochaetes, in that order. While the chironomid fauna, excepting the absence of the 'Orthocladiinae B', was nearly identical to that of the sandy bed of the Athabasca River, the dominance of oligochaetes is very different. It is interesting to note that Shadin reported that the greatest numbers of oligochaetes occurred about 10 cm below the surface of the sand and this may account, in part, for the relative absence of worms in the samples of the present study which were probably taken to a depth of only about 5 cm.

If large populations of worms exist deep in the sand and were missed by the sampling technique, the total standing stocks of invertebrates in the Athabasca River would often be much higher than the $9500 \cdot m^{-2}$ reported for the Volga River (Shadin 1956). Densities of 'Orthocladiinae B' alone were often found to be from 50 to 80% of that value in June and July. However, if oligochaetes were distributed in the sand as Shadin described for the Volga River, it seems likely that they would never have contributed more than 2% of the fauna in any collection.

Regardless of this discrepancy, it is clear, from the limited number of abundant species found on shifting river sand, that this is an unstable environment but one which can support large populations of specialized organisms. Among the best adapted and cosmopolitan of these are larvae of several genera in the *Harnischia-complex*. Saether (1977) characterized the immature stages of *Chernovskiia*, '*Cryptochironomus*' cf. *rolli*, *Beckiella tethys*, and *Robackia* spp. as inhabitants of the sandy beds of large rivers and occasionally lakes in eastern Europe, Asia, the United States, and possibly (*Beckiella* sp.) Africa. *Robackia demeijerei* has been reported from the Mackenzie drainage in the Yukon and the Northwest Territories (Wiens et al. 1975) to as far south as Florida (Saether 1977). The relatively few records of all of these species probably reflect the limited sampling effort which has been expended on large sandy rivers. Their cosmopolitan distributions emphasize the importance of the physical characteristics of shifting sand as the primary environmental factor controlling the fauna in such habitats.

In light of these observations, the lack of previous records of 'Orthocladiinae B' is somewhat surprising since it is obviously a very successful species on shifting substrates. This species was found only in the Athabasca River and one of its tributary streams, Eymundson Creek, the bed of which consists of a loose matrix of silt, gravel, and particles of oil sand. The absence of previous records is probably the result of the small size of the larvae (fourth instar larvae are less than 1.9 mm in length and about 0.2 mm in diameter) which would allow it to pass through the nets and sieves commonly employed for benthic studies. 'Orthocladiinae B' probably feeds on bacteria, protozoa, and micrometazoa among or attached to sand grains and, in turn, appears to be the primary prey of *Robackia* larvae. Recognizable remains of 'Orthocladiinae B' were found in the guts of about 10% of 50 larvae of each species of *Robackia*. The feeding behaviour of the two species may be different, however, since *R. claviger* appeared not to swallow the head capsule of its prey.

While these specialized Chironomidae were numerically dominant on sandy substrates, a variety of other organisms were also found, especially as the current regime stabilized in September and October. The most abundant of these were the stoneflies *Isoperla longiseta* and *Hastaperla brevis* which first appeared as early-instar nymphs at Sites 1 and 6 in July. Observations suggest that these and other species spread from stable rocky substrates onto gravel and coarse sand where they probably over-winter before moving back to rocky areas and accumulations of debris in the spring.

The most unexpected members of the sand fauna were the larvae and pupae of *Simulium arcticum* which averaged about $25 \cdot m^{-2}$ at Sites 1 to 5 in June and July. This species is abundant in the rapids upstream of Fort McMurray and has been reported to be a pest of cattle in that area (Fredeen 1976). Attempts have been made to control economically significant outbreaks of *S. arcticum* in 1974, 1975, and 1976, by injecting methoxychlor upstream of suspected areas of preferred larval habitat. While the numbers of simulids appear to be small on shifting sand, they amount to populations of $5 \times 10^6 . km^{-1}$

if the width of the channel is assumed to be only 200 m. Most of the river in the study area is much wider than this. This is an extremely atypical habitat for simulid larvae, which are usually considered as classical examples of attached filter feeders, and is obviously not a preferred habitat since larval densities were several orders of magnitude higher on stationary debris. (Surprisingly, larvae were never abundant on bedrock, even in June and July.) The possibility that these sand-dwelling larvae are predaceous on chironomids (Peterson and Davies 1960) should be investigated. The successful development of large numbers of *S. arcticum* within the substrate where they would be less susceptible to insecticides (Flannagan et al. in prep.) has serious implications for efforts to control nuisance outbreaks.

Another animal which deserves special mention is the mayfly *Analetris eximia*. This species did not appear in the airlift or grab samples but was regularly taken by rapidly dragging a wide-mouthed dip net over fine or muddy sand (Edmunds and Koss 1972; Lehmkuhl 1976). This species has previously been reported only from a short section of the Black Forks River in Wyoming and parts of the Saskatchewan River in Saskatchewan (Edmunds et al. 1976). It is encouraging to add the Athabasca River to the extremely short list of known habitats, but even here this rather rare species may be threatened by industrial development and insecticides (Lehmkuhl 1976).

The development of oil sand related industry along the lower Athabasca River poses problems of water quality monitoring and the enforcement of standards for municipal and industrial effluents. While benthic invertebrates have been widely accepted as useful indicators of water quality (Hynes 1965; Mason et al. 1973), the physical nature of the Athabasca River requires the use of special techniques for the collection of reproducible samples of communities of organisms sensitive to pollution. A variety of types of artificial substrates and mooring techniques have been recommended for use in large rivers (Crossman and Cairns 1974; Mason et al. 1973; Hestor and Dendy 1962; Anderson and Mason 1968;

Benfield et al. 1974; Gale and Thompson 1974; Mason et al. 1973; McCart et al. 1977; Flannagan et al. in prep.). However, the results of the present study cast serious doubt on the reliability of conclusions drawn from the colonization of artificial substrates in the Athabasca River. Any solid object placed in the river tends to collect large quantities of debris (sticks, leaves, grass, etc.), especially during periods of high water. The rocks in baskets supported few, if any, animals, even after immersion for nearly three months. However, stones from the same source were heavily colonized in the Muskeg River (Barton and Wallace in prep.). The debris contained a rich community which was different in composition from that found directly on the bottom of the river. These large, apparently pollution-sensitive species have great appeal as indicators of water quality (McCart et al. 1977), but it has been shown that large numbers of these same species can drift for very long distances and rapidly recolonize denuded areas of the Athabasca River (Flannagan et al. in prep.). Since the quantity of debris and the proportion of the various species in the drift depend on the discharge of the river and the season, the composition and abundance of invertebrates on artificial substrates in the lower Athabasca River probably does not reflect local water quality.

Direct sampling along the water's edge using standard grabs or Surber samplers is also of doubtful reliability, especially for comparing sites over time. Fluctuations in water level have been shown to have a deleterious effect on macrobenthic communities (Hynes 1961b; Hunt and Jones 1972), and the results clearly showed that the continual erosion and deposition of fine sediments lead to substantial changes in the abundance and species composition of the fauna. During late summer and autumn, when the river is low, carefully selected sites might be compared using conventional techniques, but further research is needed to assess the short-term effects of siltation and fluctuations of the water level on benthic communities of large rivers.

Until such studies are done, sampling sites for monitoring purposes should be located away from the shore and on substrates which are consistent in texture over long periods of time, such as bedrock and the coarse sand and gravel of the main channel. The airlift used in the present study has the advantage of collecting fairly reproducible samples from the entire range of substrates, but these are not easily quantifiable except on sand. A pole-mounted grab with an extremely strong closing mechanism might be preferable for loose sediments. When the river is free of ice and large objects floating downstream, divers could collect samples using the airlift and a quadrat (Barton and Hynes 1978) or a Surber or Hess sampler (Section 2; also Rabeni and Gibbs 1978; Gale and Thompson 1974). Obviously, no single technique is best for all substrates at all times. Interpretation of results will be difficult, also, until further research reveals the range of variations in standing stocks, both temporally and spatially, and the sensitivity of the psammophilic fauna to expected effluents. Simple indices of water quality, such as number of species or diversity, do not appear to reflect the ecological health of this limited community.

For these reasons, artificial substrates may, after all, be the most versatile technique for the biological assessment of water quality. However, it is essential that these be shielded in some way to prevent the accumulation of debris. Also, rock-filled baskets would probably give better results if the stones were incubated in a tributary stream to develop a film of aufwuchs before being placed in the Athabasca River.

3.2 STANDING STOCK OF MICRO- AND MACROBENTHOS IN THE ATHABASCA RIVER IN AUTUMN

3.2.1 Introduction

This section presents quantitative estimates of the standing stocks of microbenthos on bedrock and macrobenthos on the entire range of sediments in the Athabasca River in autumn.

Relationships between these two broad categories of organisms are considered with respect to discharge and substrate during the season when the fauna is approaching the relatively stable conditions of winter.

As mentioned in a preceding section (2.3), the sampling technique used in the monthly comparison of the principal study sites on the Athabasca River did not provide quantitative information about the organisms inhabiting rock substrates. Further, information regarding the importance of benthic algae and bacteria on substrates utilized was not gathered at that time. In late September, as discharge was declining, it became possible to obtain such data through diving. Samples of macro- and microbenthos were collected along a transect oriented perpendicularly to the shore on exposed bedrock at Site 6. The transect was continued on the opposite side of the river to provide comparable data on the macrofauna of sandy substrates. To complete the description of the fauna of the entire range of river sediments, samples collected from sand and mud in early October 1976 were also examined.

3.2.2 Methods

Macroinvertebrates on sand and mud substrates were sampled using a 15 cm^2 Ekman grab. The device was forced into the substrate by hand and, if necessary, the jaws were kicked shut before the grab was lifted from the bottom of the river. On 7 October 1976, 34 samples were collected from depths of 0.1 to 1.1 m along a large, exposed sandbar. Ten samples were taken from coarse sand at the upstream end of the bar, six from medium sand off the middle, nine from fine sand at the downstream end, and nine from muddy sand and mud from the backeddy behind the bar.

Samples of benthic invertebrates from rocky substrates were collected on 27 September 1977 by a diver using a 202 µm Surber sampler (0.09 m^2). Lead weights (2 kg each) were attached to the sides of the sampler so that it would remain in place in currents up to 1 $m \cdot s^{-1}$. Both diver and Surber sampler were suspended on freely sliding rings around a line which was tied to a boulder on shore at one end and to a heavy anchor at the other. The diver located the sampling sites using a wrist depth gauge. The Surber was placed firmly on the river bed, any large, enclosed stones were lifted and scrubbed in front of the net, and the remaining sediments were thoroughly agitated to dislodge benthic animals.

Three samples each were collected from rubble and bedrock at depths of 0.2 m, 1.5 m, 3.0 m, and 4.5 m on 27 September 1977. Three samples were taken with the Ekman grab from sand on the opposite side of the river at depths of 0.2 m and 1.5 m. One additional sample was collected with the weighted Surber from very coarse sand and gravel at a depth of 4.5 m.

The nature of the substrate from which each sample was taken was determined visually. The types of substrates recognized included bedrock and rubble (particles >10 cm), gravel (1 to 10 cm), pea gravel (3 to 10 mm), very coarse sand (1 to 3 mm), coarse sand (0.5 to 1.0 mm), medium sand (0.3 to 0.5 mm), fine sand (<0.2 mm), and mud.

Samples collected by either method were emptied into a bucket and the organisms were concentrated from them by adding river water, agitating, and decanting through a 180 μm sieve. This process was repeated eight times or until no new organisms appeared on the sieve after three successive decants. The sediments remaining in the bucket were examined for large molluscs and heavily cased caddis larvae which were added to the material on the sieve. All samples were preserved with 10% formalin in the field and organisms were separated from the remaining debris under 10X magnification and stored in 70% alcohol. (The possibility exists that not all organisms were eluted from the samples by this technique, but the relatively small variation between replicate samples suggests that the technique was at least consistent.)

Animals were identified and enumerated at the lowest possible taxonomic level, usually generic or specific. Chironomidae were verified by comparison with specimens in the Canadian National Collection (Ottawa). Trichoptera were confirmed by R. Crowther

(University of Calgary). After counting, each sample was filtered and the organisms on the filter paper were allowed to air dry for 30 minutes at room temperature and weighed to the nearest 0.1 mg.

A sterile water sample was collected from 15 cm above the substrate of each depth along the rubble bottom transect for the enumeration of suspended bacteria. Immediately after collection, five standard (Jones and Simon 1975) 3 mL subsamples (of each sample) were filtered to dryness through sterile, 25 mm, 0.2 μm Nuclepore filters using a sterile Millipore filtration apparatus. Each filter was then lightly attached to a Millipore media pad in a sterile petri dish by two small spots of rubber cement applied to the periphery. Two to three drops of 0.5% glutaraldehyde on a small ball of cotton wool were added to fix and preserve each sample. On return to the Mildred Lake laboratory, the filters were stained with 0.01% Acridine Orange in potassium phosphate buffer for 2 min and then destained with iso-propanal in a Millipore filtration apparatus. The stained membranes were air dried, mounted on a glass slide containing a drop of immersion oil (Cargille Laboratories), and covered with a coverslip (Geesey and Costerton in press). The membranes were viewed with a Leitz Ortholux II microscope fitted with a Fluorescence vertical illuminator with a flat field objective at a magnification of 1250X. Illumination was provided by an Osram ultra high pressure mercury lamp (200W/4) with an excitation filter setting of 5 and a suppression filter setting of 3. Bacteria fluorescing orange or green were counted in 10 different 0.0047 mm^2 fields. In general, 20 to 100 cells were counted on each filter.

Enumeration of sessile bacteria was based upon the method of Geesey and Costerton (in press) and involved removing a 4 cm^2 area of periphyton using a template and sterile scalpel from five individual rocks collected by the diver from each sampling depth. Detached material was transferred to sterile vials containing 10 mL of freshly prepared sterile 0.5% glutaraldehyde (EM grade, Ladd Industries) in 0.067 M cacodylate buffer (Sigma Chemical Company) equilibrated to river temperature. On return to the laboratory, the bacteria were dispersed by blending the sample for 30 s at setting 5 on a Brinkmann Polytron PCU-2 homogenizer using a PT-10 generator. Initial tests had demonstrated that this was the shortest time permitting maximum dispersion of bacteria. Cleaning of the probe between samples was achieved by running the generator at maximum speed in 10 mL of cacodylate buffer for 90 s. Following appropriate dilution, a standard volume (Jones and Simon 1975) of 3 mL of the suspension was filtered onto sterile 0.2 µm Nuclepore filters and stained and viewed as above. All the solutions had been filtered, sterilized, and, in the case of the buffers, autoclaved. Periodic checks for contamination at all stages yielded negative results.

Determination of ATP biomass involved the on-site aseptic removal of 4 cm^2 areas of periphyton from five separate rocks and immediate extraction of ATP with 4 mL of boiling Tris [Tris - (hydroxymethyl) aminomethane] buffer (Holm-Hansen and Booth 1966). Samples were frozen on return to the field laboratory, approximately 1 h later. ATP was determined using the Luciferin-Luciferase assay. Approximately 12 h prior to assaying for ATP, a crude freeze-dried extract of luciferin-luciferase from firefly lanterns (Sigma FLE-50) was reconstituted with water and held at $5^{o}C$. Immediately prior to assaying, the samples were thawed, centrifuged at 15 000 RPM for 5 min, and then 0.5 mL of the supernatant was added to 2.0 mL of arsenate buffer (50 mmol solium arsenate, 5 mmol potassium phosphate, 5 mmol magnesium chloride - pH 7.8). The reaction was initiated by the addition of 0.1 mL of the firefly lantern extract, vortexed for 5 s, and the photon emission counted exactly 60 s later over a 0.1 min period in an Isocap 300 scintillation counter set for tritium with the photo-multiplier tube "out of coincidence" (Stanley and Williams 1969). All solutions were equilibrated to $0^{o}C$ in an ice-bath. A standard curve was produced each time using equine muscle ATP (Sigma FF-ATP). All glassware was acid-washed before use and rinsed three times with distilled water.

Chlorophyll *a* was determined by scraping 4 cm^2 areas of periphyton from five separate rocks from each depth and transferring these to vials containing filtered river water. The contents were filtered through a Whatman GRC fibreglass filter and magnesium carbonate was sprinkled onto the retained material. The filters were then folded and wrapped in aluminum foil and frozen on return to the field laboratory. Pigments were later extracted using 15 mL of 90% acetone, homogenized for 30 s in a Brinkmann Polytron PCU-2 at maximum setting, and allowed to stand at -18°C in the dark for 24 h prior to determining chlorophyll *a* using the method of Moss (1967a, 1967b).

All frozen material stored in the Mildred Lake field laboratory was transferred to the Edmonton laboratory in a frozen state using an insulated box containing dry-ice.

Photosynthetically active radiation (400 to 700 nm), PAR, was determined at several depths using a Li-Cor quantum sensor (Li-192S) coupled to a Li-Cor quantum meter (Ll-18S).

3.2.3 Results

On both sampling dates, the temperature of the Athabasca River was about 12°C and the discharge was about 800 $m^3 \cdot s^{-1}$. The transparency of the water was increasing after very turbid conditions during high summer discharge, but light levels declined rapidly with depth. At 0.25 m in 1977, PAR was 45% of the value at the surface while the 1% light level occurred at 2.0 m and the 0.01% level at 3.0 m. Numbers of suspended bacteria collected from water 15 cm above the rubble substrates ranged from $20 \times 10^5 \cdot mL^{-1}$ at a depth of 3 m, to $2.6 \times 10^5 \cdot mL^{-1}$ at 4.5 m (Figure 27), but none of the values were significantly ($p > 0.05$) different from the others.

Greatest numbers of sessile bacteria were found at 0.25 m and 3.0 m: $1.9 \pm 10^7 \cdot cm^{-2}$ (95% confidence limits) and $1.4 \pm 0.25 \times 10^7 \cdot cm^{-2}$, respectively (Figure 27). The former was associated with a thick (ca. 3 mm) layer of loose silt. The latter was associated with a thin (ca. 1 mm), adherent film which appeared

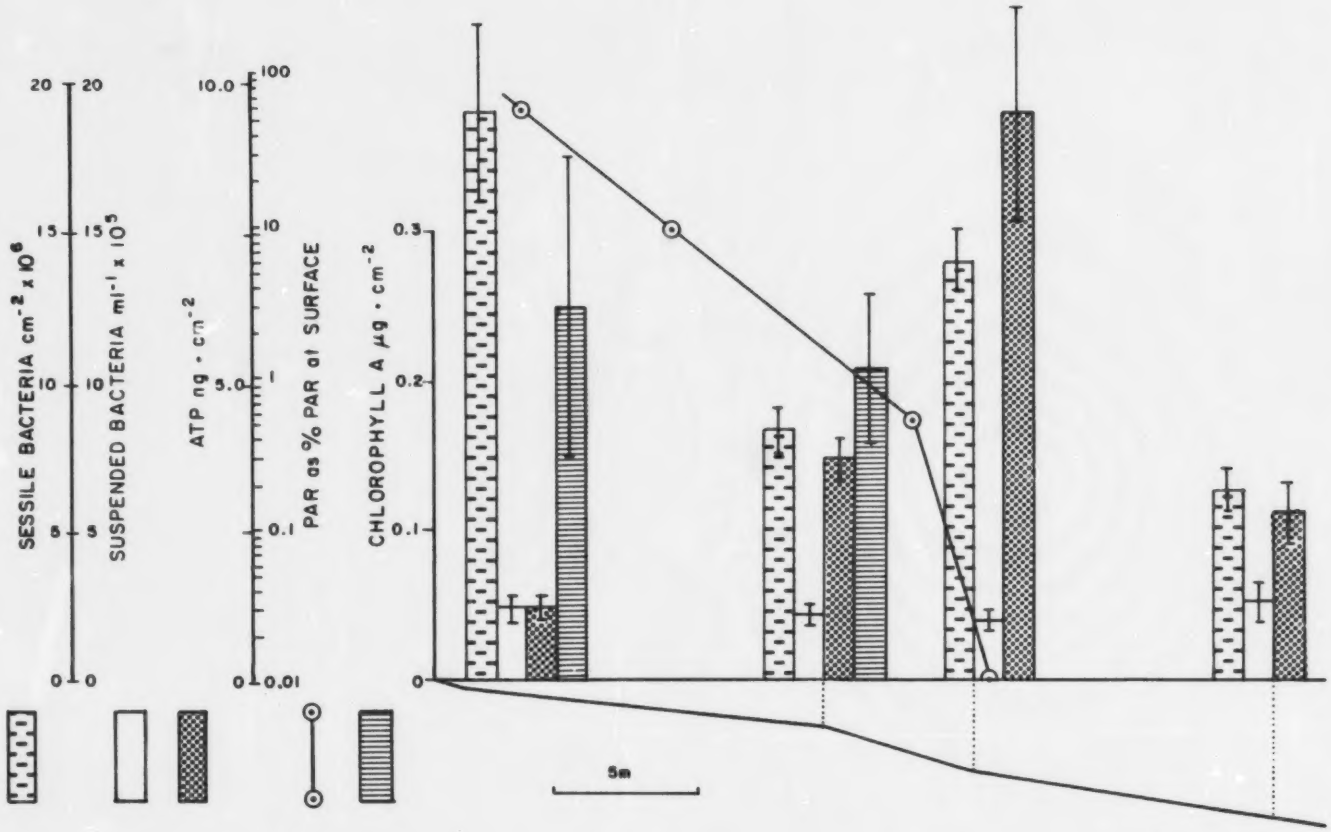

Figure 27. Light penetration and standing stocks of sessile bacteria, suspended bacteria, ATP, and chlorophyll a along the bedrock transect. PAR means photosynthetically active radiation.

to be similar to that found also at 1.5 and 4.5 m. While the mean numbers of sessile bacteria at 0.25 and 3.0 m were not significantly different from each other, both were significantly greater than the means at 1.5 and 4.5 m ($t = 2.68$ to 10.40; 8 df).

The amount of sessile ATP increased steadily with depth down to 3.0 m, but then dropped at 4.5 m (Figure 27). The amount of chlorophyll *a* was similar at both 0.25 and 1.5 m, the only depths at which measurable quantities were found (Figure 27).

The variety of benthic invertebrates differed among the five substrates which were sampled. Rubble and bedrock yielded a total of 56 taxa; mud, 18; coarse sand, 15; fine sand, 12; and medium sand, 9 (Table 18).

The numerical density and total invertebrate biomass on rocky substrates exhibited patterns of depth distribution similar to those of sessile ATP and sessile bacteria, respectively (Figure 28). Though slightly more taxa were also found at 3.0 m (a total of 37 from the three samples) than at 0.25 m (31), 1.5 m (33), or 4.5 m (30), the mean number of taxa per sample was about 20 at all depths. The large biomass at the water's edge was mainly due to the presence of the corixids, *Callicorixa audeni* and *Sigara solensis*, at a density of about 70 individuals$\cdot m^{-2}$. If these and other organisms greater than 6 mm in length are excluded, biomass followed a pattern similar to that of numbers of animals. The species composition of the fauna at 0.25 m was also somewhat unique (Table 18), probably due to the fairly heavy layer of silt which covered the stones there.

The standing stock of benthic invertebrates on sand and mud were much lower than on rock in terms of biomass, but not in numbers of individuals (Figures 28 and 29). While larger insects such as Ephemeroptera, Plecoptera, and certain Diptera (especially Empididae) numerically dominated the fauna on rocky substrates, Chironomidae were the most abundant group in finer sediments. The series of samples taken in 1976 along a continuum of sediments of decreasing mean particle size showed a distinct change in the composition and density of the benthic community on sand where the

Table 18. Invertebrate taxa found on bedrock and rubble (R), coarse sand (C), medium sand (MS), fine sand (F), and mud (M).

Taxa	Substrate[a]				
	R	C	MS	F	M
Hydra	+[a]	+	+	+	+
Nematoda	+	+	+	+	+
Enchytraeidae	+				+
Tubificidae	+				+
Amphichaeta	+				
Nais behningi	+			+	
Copepoda	+				
Ostracoda		+	+		+
Acari	+[c]				
Baetis	+[b]				
Ameletus	+[b]				
Ametropus neavei	+[b]		+	+	
Analetris eximia	+[b]				
Ephemerella inermis	+				
Leptophlebia	+[b]				
Heptagenia	+				
Rhithrogena	+				
Ophiogomphus columbrinus	+				
Oenopteryx	+				+
Acroneuria abnormis	+				
Isogenoides frontalis	+	+			
Isoperla longiseta	+				
Hastaperla brevis	+				
Neotrichia	+[c]				
Cheumatopsyche	+[c]				
Hydropsyche	+[c]				
Oecetis	+[c]				

continued ...

Table 18. Continued.

Taxa	R	C	MS	F	M
Brachycentrus	+[c]				
Callicorixa audeni	+	+			
Sigara solensis	+				
Optioservus fastiditus	+				
Eriocera	+				
Ceratopogonidae	+				
Simulium	+[c]				
Atherix pachypus	+				
Empididae	+				+
Ephydridae			+		+
Larsia	+				
Procladius	+				+
Thienemannimyia-gp.	+				
Ablabesmyia		+			
Monodiamesa	+				
Chernovskiia orbicus			+		
Chironomus fluviatilis-gp.					+
Cryptochironomus				+	
Paracladopelma spp.	+[b]	+	+	+	+
Polypedilum sp.	+				
P. breviantennatum-gp.	+[b]	+	+	+	
P. fallax-gp.	+				
Robackia claviger	+[c]	+		+	+
Stenochironomus	+				
Stictochironomus	+				
Cladotanytarsus	+[c]				
Micropsectra				+	+
Rheotanytarsus	+	+	+	+	
Stempellina					+
Tanytarsus	+	+			

continued ...

Table 18. Concluded.

Taxa	R	C	MS	F	M
Corynoneura	+				
Thienemanniella		+			
Cricotopus		+			+
Eukiefferiella	+				
Heterotrissocladius	+				
Nanocladius ?rectinervis	+			+	+
Parakiefferiella	+				
Orthocladiinae B	+	+	+	+	+

[a] Present

[b] Taxa found only at 0.25 m

[c] Taxa not found at 0.25 m

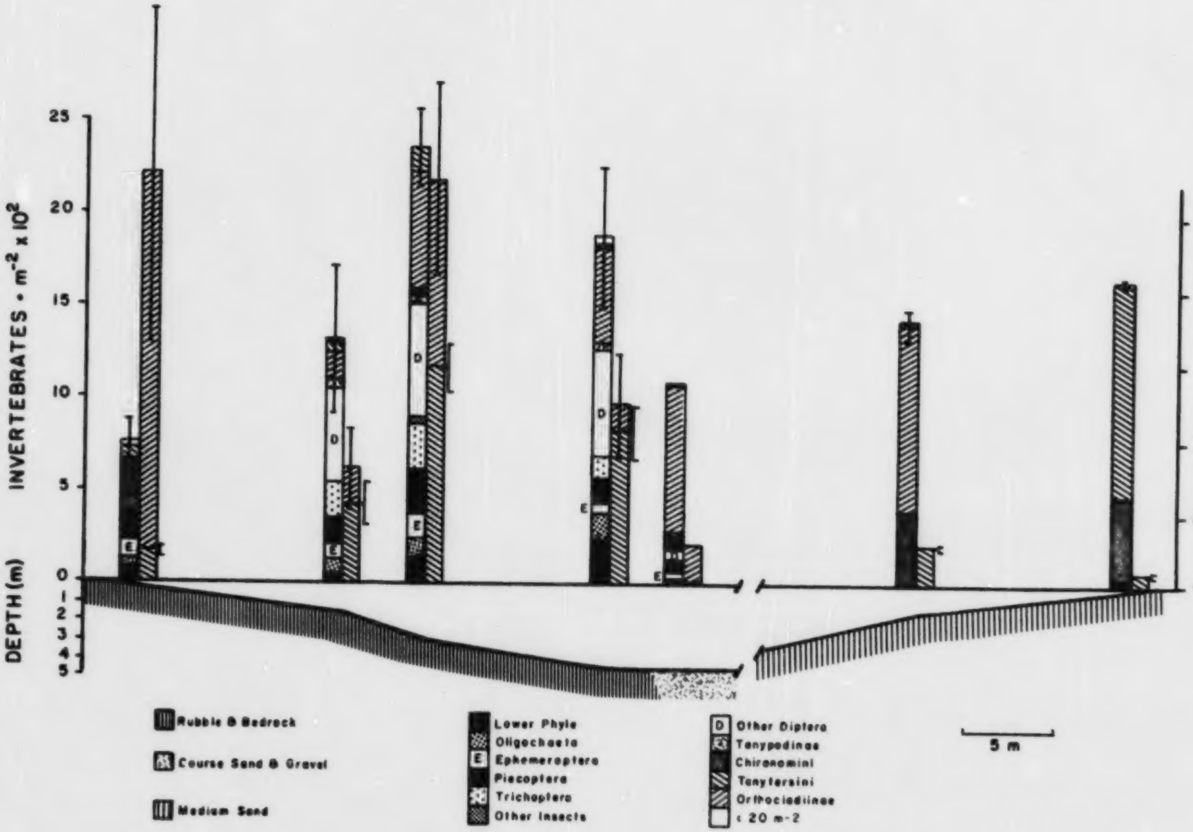

Figure 28. Mean (± 1 S.E.) density and biomass (right hand bar of each pair) of invertebrates found in the 1977 transect. The lower portion of each biomass bar represents animals less than 6 mm in length.

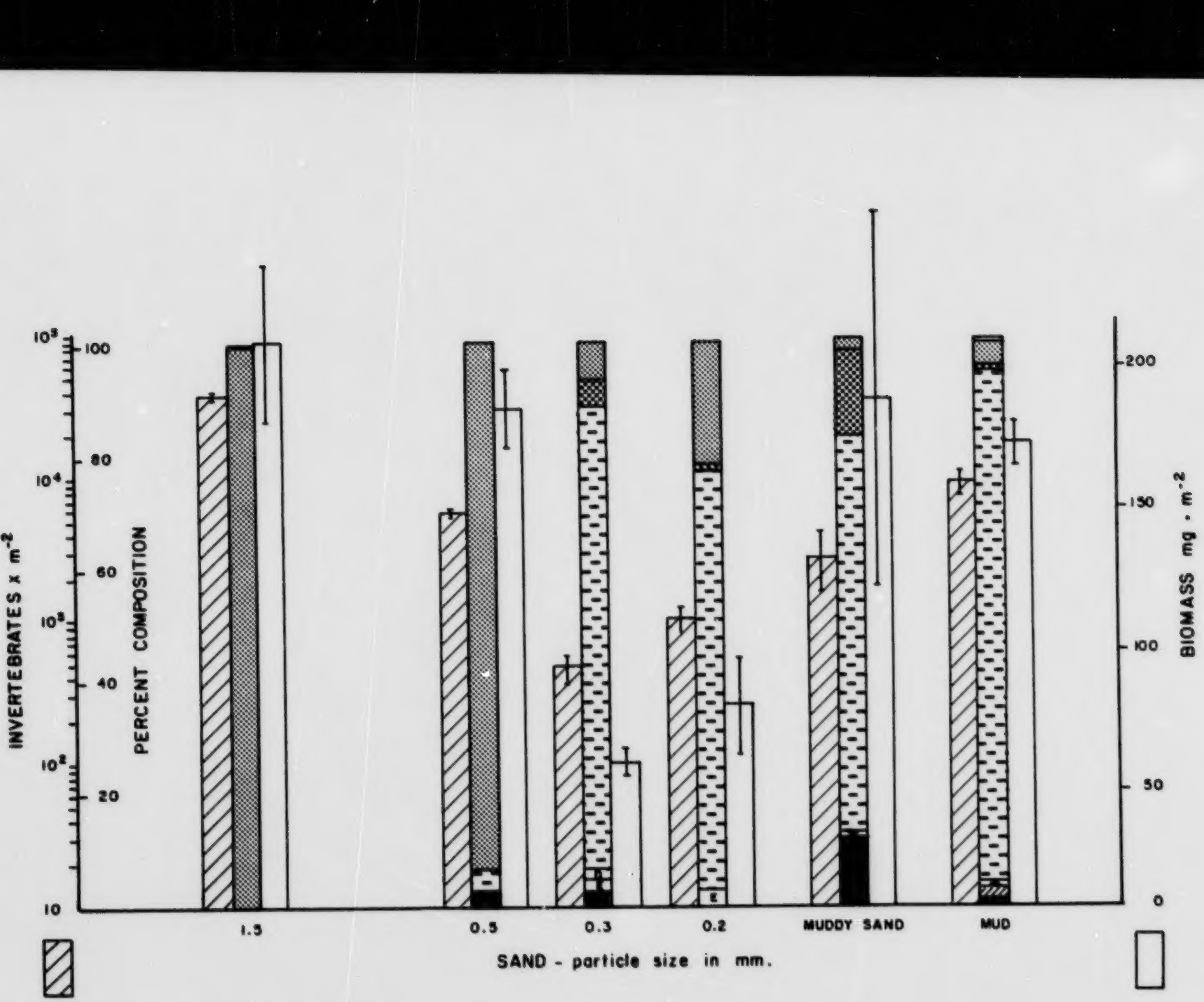

Figure 29. Density (± 1 S.E.), percent composition, and biomass (± 1 S.E.) of invertebrates on sand and mud, 1976.

mean particle size was about 0.3 mm (Figure 27). On coarser sand, 'Orthocladiinae B' was overwhelmingly abundant. On finer sediments, a variety of Chironominae became increasingly more important. Despite distinct differences in numerical standing stocks (39 000·m^{-2} on very coarse sand versus 8300 on mud), biomass was essentially the same at both ends of the sediment range. This was due to the very small size of 'Orthocladiinae B', the fourth instar larvae of which are less than 2 mm in length.

Since both the Surber and Ekman samplers were rarely used, in this study, in the manner for which they had originally been designed, it was somewhat surprising that the sampling efficiencies of both devices were the same. The standard error expressed as a percentage of the mean number of individuals in the samples ranged from 9 to 30% for the weighted Surber, and from 3 to 25% for the Ekman.

3.2.4 Discussion

The results of this study emphasize some basic similarities between benthic communities of rocky substrates in running waters, regardless of the size of the stream or river or its geographical location. Certain groups of macroinvertebrates, such as the Heptageniidae, Hydropsychidae, Simuliidae, some Baetidae, and Plecoptera (or their ecological equivalents), are characteristic of rocky river beds throughout the world (Hynes 1970; Elgmork and Saether 1970; Allen 1951; Behning 1928), though the relative contribution of each to the total fauna may vary from place to place and with season. Quantitative information on standing stocks of both macro- and microbenthic organisms on bedrock is extremely limited. It appears that where the thickness of the inhabitable substrate is limited to a few centimetres or less, standing stocks of macroinvertebrates in autumn average about 2000 individuals·m^{-2} in the Athabasca River as well as in other streams (Armitage 1958; Pennak and Van Gerpen 1947; Rabeni and Gibbs 1978--assuming a loss of about 50% of numbers due to the large mesh collecting nets used in three studies) and large lakes (Barton and Hynes 1978).

Standing stocks of bacteria on rocky river beds also seem to fall within a fairly narrow range. Geesey et al. (in prep.) reported 9.0×10^6 to 1.0×10^8 bacteria$\cdot$cm^{-2} in three small subalpine streams during October. In two streams tributary to the Athabasca River near the study area, Lock et al. (in prep.) found 1.4×10^7 to 1.3×10^8 bacteria$\cdot$cm^{-2} during late September. The range found in the present study, 8.4×10^6 to 1.9×10^7, was slightly lower than in these smaller streams but considerably higher than the values of 9.4×10^4 to 4.5×10^5 bacteria$\cdot$cm^{-2} reported by Sládecek and Míshlovský (1976). Sládecek and Míshlovský examined bacteria growing on glass slides which had been submerged in a river for two days. It seems reasonable to conclude that exposure for two days is insufficient for the development of a stable microbial community and that the results obtained in this manner probably do not reflect the actual densities to be found in the river.

The levels of ATP on rocks in the Athabasca River ranged from 1.2 to 9.45 ng$\cdot$cm^{-2}, values considerably lower than the 4 to 400 ng$\cdot$cm^{-2} reported by Geesey et al. (in prep.) and the 24.9 to 25.4 ng$\cdot$cm^{-2} in tributaries of the Athabasca River (Lock et al. in prep.). In all of these studies, boiling Tris buffer was used as the ATP extractant because of its simplicity. It is likely that the extraction efficiency of this technique was low (Lee et al. 1971; Karl and La Rock 1975; Bancroft et al. 1976) and neither was it possible to determine the efficiency by incorporation of an internal bacteria ATP standard (Bancroft et al. 1976; Jones and Simon 1977) because of the remoteness of the field site. Since the intention was to use the level of ATP as an index of living biomass, it was assumed that the Tris extraction method would allow comparison of relative levels from similar substrates.

A poor correspondence was found between estimates of ATP and microbial biomass as indicated by chlorophyll *a* and direct counts of bacteria. However, lowest ATP levels were associated with the lowest biomass of small invertebrates (<6 mm) and conversely the highest ATP levels occurred with the highest biomass of small invertebrates, many of which were small Tanytarsini and Empididae likely to have been included in the scraped material used for the ATP extractions. This phenomenon is also evident in the studies of Geesey et al. (in prep.) and Lock et al. (in prep.). In the latter study, there is an indication that the ATP may be primarily of animal origin, i.e., Protozoa, micro-metazoa, Bryozoa, etc.

The chironomid-dominated macrobenthic communities found on less stable substrates, mud and sand, in the Athabasca River were somewhat different from those which have been reported elsewhere. Previous studies of large rivers, such as the Athabasca River (McCart et al. 1977), Volga River (Shadin 1956), and the Ural River (Zachetnova 1975), have shown oligochaetes to be the dominant organisms on mud and, to a lesser degree, on sand. This group contributed only a small number to the total fauna in the Athabasca River. This is probably not due to the season at which they were collected since studies of oligochaete production (Johnson and Brinkhurst 1971b, Jonasson and Thorhauge 1972) indicate that standing stocks should be near their highest levels in early autumn. Since the Athabasca River in the AOSERP study area is confined by steep banks, the fluctuating discharge during the open-water season causes mud deposits to be very transitory along the main channel of the river. This may prevent the development of large populations of oligochaetes. Many Chironomidae, especially certain Chironomini, appear to have somewhat broader substrate tolerances and there is evidence that they are able to move rapidly into newly available areas (Moon 1940; Darby 1962; McLachlan 1969).

Most of the bottom of the Athabasca River in the AOSERP study area is sand which is deposited in different size fractions according to localized differences in current velocity. A small group of chironomids, including *Polypedilum breviantennatum*-gp. *Cryptochironomus*, *Robackia claviger*, and *Paracladopelma* spp., dominated the fauna of fine and medium sand. These same species or species-groups have been found on similar substrates in rivers in Russia (Shadin 1956) and the southeastern United States and larger lakes (Saether 1977). Shadin (1956) pointed out that all are predaceous, elongate forms with thick cuticles, well-adapted to burrowing in unstable sands.

Extensive deposits of medium and fine sands occur in the Athabasca River only out of the main current--along sandbars or the shore. The bed of the main channel in the study area consists of very coarse sand which supported a macrofauna composed almost exclusively of one species of chironomid, 'Orthocladiinae B'. This species has never been reported before, probably due to its small size (fourth instar larvae are about 1.9 mm long and 0.2 mm in diameter) and its preference for coarse river sand. It was found only in the Athabasca River and Eymundsen Creek, despite extensive sampling in other tributary streams in the area.

An average of about 40 000 organisms$\cdot m^{-2}$ was found on very coarse sand in 1976, but due to the small size of individual larvae, total biomass (0.2 $g\cdot m^{-2}$) was the same as on mud and muddy sand which supported only about 8000 and 2700$\cdot m^{-2}$, respectively. Much higher estimates of biomass were given by Shadin (1956) for the sandy bed of the Volga River: 1.2 $g\cdot m^{-2}$ and 9500 individuals$\cdot m^{-2}$, consisting mostly of oligochaetes and amphipods. Berner (1951) estimated the mean standing stock on fine sand in the lower Missouri River at 0.07$\cdot m^{-2}$, but concluded that the coarse shifting sand in the main channel probably did not produce any benthic organisms.

It is significant to note that amphipods were not found in the samples, nor were any reported by Berner (1951). *Pontogammarus*, the most abundant amphipod on sand in the middle and lower Volga River (Shadin 1956), is restricted to the Caspian drainage. There seem to be no records of amphipods living on sifting river sand in North America (Bousefield 1958; Holsinger 1972). Clearly, the sandy beds of large rivers are a very severe habitat which supports a highly specialized fauna. Berner (1951) felt that allochthonous material is the principle source of energy for the benthos of large silty rivers, since primary production is largely eliminated by turbidity. In geographical areas where the appropriate, specialized organisms exist, large standing stocks of invertebrates can be supported by this allochthonous material. Where the number of specialized forms is more limited, much of the organic matter probably passes straight through the system.

Autochthonous primary production does contribute organic matter to large rivers despite the rapid attenuation of light with depth in turbid water (Ertl and Tomajka 1973). In a summer study of the distribution of chlorophyll *a* with depth in the Danube, Ertl et al. (1975) found 17% of the surface standing stock occurring at 2 m. In the Athabasca River, the standing crop at 1.5 m was about 50% of that at 0.25 m. The light level at 1.5 m was approximately 1% of the surface, the generally accepted compensation-point for algae, while at 0.25 m, the light was 45% of that at the surface. The turbidity of the Athabasca River was 6.45 J.u. and chlorophyll *a* averaged about 0.2 $\mu g \cdot cm^{-2}$. Since the standing stock of chlorophyll *a* in the nearby Steepbank River, at a depth of ca. 20 cm, was about four times as great (0.8 $\mu g \cdot cm^{-2}$) at similar levels of turbidity (6.65 J.u.) (Lock et al. in prep.), it seems likely that the low levels of chlorophyll *a* at 0.25 m in the Athabasca River were due to factors other than light, such as fluctuation of the water level or deposition of silt. It should be emphasized that these comparisons are based upon standing crop data and the potential for primary production of the river remains to be determined. The deposition of silt along the water's edge is probably also responsible for the low numbers of macroinvertebrates found at 0.25 m on rock (Hynes 1961b; Hunt and Jones 1972; Rosenberg and Snow 1975).

Considering the fact that the turbidity of the Athabasca River in September was at its lowest level of the 1977 open-water season, the standing stock of benthic primary producers (as indicated by chlorophyll *a*) was probably at its highest. The poor correspondence between chlorophyll *a* and numbers of invertebrates suggests that benthic algae are less important as a food source than are other dietary components (Moore 1977). The density of invertebrates on rock substrates did correspond closely with the density of bacteria. This may represent a direct response to a food source (Brinkhurst and Chau 1969; Cummins 1973; Fredeen 1960), although Baker and Bradnam (1976) recently suggested that the role of bacteria in the nutrition of aquatic detritovores may have been overestimated. However, since the greatest densities of benthic animals were found well below the compensation-point for algae, it seems reasonable to conclude that the principle source of energy for the invertebrate community was allochthonous in nature.

Finally, it should be emphasized that, on rock, both the micro- and macrofauna were most abundant at the depth where conditions were probably most stable, namely 3 m. Lower densities at 4.5 m were probably the result of periodic scouring by sand caused by changes in the discharge of the river. Near the water's edge, alternating siltation and erosion would likewise reduce the fauna (Hynes 1961b; Hunt and Jones 1972; Shadin 1956).

4. EXPERIMENTAL MANIPULATIONS

4.1 THE EFFECTS OF SYNTHETIC CRUDE OIL ON BENTHIC COMMUNITIES UNDER SUNLIT AND DARK CONDITIONS

4.1.1 Introduction

The industrial development of the Athabasca Oil Sands region may increase the probability of disturbances to aquatic habitats, especially diversions of rivers and the discharge of industrial or municipal effluents.

Studies in this section were aimed at elucidating the responses of freshwater bacterial, algal, and macroinvertebrate communities to contamination of substrates by oils.

The Great Canadian Oil Sands Limited (GCOS) plant provided synthetic crude oil and its components for experiments which essentially simulated a massive, short-term spillage of such an oil into a brown-water stream. Previous freshwater toxicological invertigations of oily effluents have centred upon the effects on test species of fish (Pessah et al. 1973; Côté 1973) or upon a chemical characterization of such effluents (Hrudey 1975). While such work is useful for regulatory or enforcement purposes, it does not address the substantive issues about the assimilative capacities of contaminated streams or of the effects of such chemicals on the benthic communities which ultimately determine the productivity of such rivers. Smith (1974) summarized the literature on the effects of the many chemicals which may emanate from the Canadian petrochemical industry, and emphasized the complexity of the subject. As Parker et al. (1976:291) noted, "only recently have small and/or continuous low level contaminations by petroleum and petroleum products started to receive similar attention". Only very little is known about the longer term aspects of low level contamination of fresh waters.

Although the effects of petroleum contamination in fresh waters are rarely as spectacular as massive marine spills, they may be as damaging (Blumer et al. 1971; Tarzwell 1971), but are certainly poorly understood, especially for the type of brown-water streams which characterize much of the Canadian boreal forest zone. While crude oils may persist in fresh waters, processes of physical weathering or biotic activity may change the amounts or types of oils which remain. Research on the effects of hydrocarbons on benthic communities has been done principally in marine littoral areas and has concentrated on the heavier grades of oils (Parker et al. 1976; Moore and Dwyer 1974). There is considerably less information available on the impacts on benthos by deposits of oils upon sediments of running water, although there has been increasing attention paid to this area (Anon. 1977; Radcliffe and Murphy 1969; Scott and MacKay 1976).

It is known that the toxicity of crude oil to aquatic organisms depends upon many factors, including the source of the oil (Sacherer 1970) and its chemical composition. Oils of low molecular weight are generally more toxic than those of greater weight and the soluble fractions are known to be toxic to fishes and some aquatic species (Smith 1974). Parker et al. (1976) found an inverse correlation between the toxicity of oils to aquatic species and the molecular weight and the degree of emulsification. However, as was noted by Berry and Brammer (1977), comparisons of petroleum toxicant data, when derived from different laboratory methodologies, may provide little useful comparative data as factors such as inter-specific and intra-specific toxic responses may vary. For instance, Berry and Brammer (1977) found that the water-soluble fractions of gasoline are more toxic to the younger instars of *Aedes aegypti* and that they were most susceptible during moulting. As a consequence of such variabilities in the results of acute toxicity testing, Tarzwell (1969) devised rigidly standardized testing procedures for oil and oil dispersants.

It was felt that such laboratory tests would be of limited applicability to the oil sands area and the contention was that it was first necessary to investigate the effects of a gross, experimental spillage of oil in a river so as to obtain an overview of the community responses which occurred within the first six months.

While it is recognized that such field studies have many limitations, the lack of experimental field studies on the effects of oil on benthic communities of running waters made such initial research a requirement before further, more complex, studies could be done.

Studies concerning the effects of oil spills upon the micro- and macrobenthic organisms of rivers are limited in the extreme (Snow and Rosenberg 1975a, 1975b; Parker et al. 1976). Rosenberg and Wiens (1976) and others have detected various responses to oil by different invertebrate species and have hypothesized that these may have been related to an enhanced growth of algae.

The present study was designed to examine the response of a brown-water river community to an entirely new hydrocarbon mixture, synthetic crude oil, the end product of the oil sands extraction process. Specifically, the objective was to examine how limestone substrates which were contaminated with synthetic crude oil were colonized by micro- and macrobenthic organisms under two regimes of light and dark. The intention was that the communities which developed in the dark would have greatly reduced algal populations and this might, therefore, shed new information on their role in community responses to oil contamination. This, in turn, would permit more informed judgements as to the relative capacities of rivers in the oil sands study area to assimilate refined hydrocarbons.

4.1.2 Materials and Methods

This study was done in a 50 m section of the brown-water Muskeg River, about 10 km above its confluence with the Athabasca River in northeastern Alberta (57°08'N, 111°35'W) (Figures 1 and 2). The substrate at the study site consisted of gravel and limestone pebbles about 5 cm in greatest diameter. The physical-chemical characteristics of the river during the study are summarized in Table 19.

In order to provide as natural a substrate as possible for floral and faunal colonization, but at the same time to avoid complications due to small differences in surface texture of mineral

Table 19. Physical-chemical parameters of the Muskeg River during the study period.

Characteristic	Value
Mean Discharge	$4.65\ m^3 \cdot s^{-1}$
Temperature	16°
pH	8.1
Conductivity	$260\ mmoh \cdot cm^{-2}$
Dissolved Organic Carbon	$24.5\ mg \cdot L^{-1}$
$NO_3 - NO_2$ Nitrogen	$0.014\ mg \cdot L^{-1}$
NH_4 Nitrogen	$0.01\ mg \cdot L^{-1}$
PO_4 Phosphorus	$0.006\ mg \cdot L^{-1}$

composition, smooth bricks, measuring 12 x 6.3 x 6.3 cm (382 cm^2), were cut from Tyndall stone commercial limestone. The bricks were placed flat on the river bed in a uniform array on a matrix pattern (Figure 30) with the narrow end upstream and with flat side uppermost. Seven columns of five bricks each were established for each of two stations.

A 1.85 x 2.45 m wooden frame covered with four layers of opaque black polyethylene was suspended from four steel stakes over one of the stations ("dark") of bricks (Figure 30). Plastic "skirts" were hung from the sides; these extended into the water but did not distort the flow of the water beneath the shade.

Measurements taken with a Licor Ll-185 quantum light meter with an underwater quantum sensor Ll 1925 indicated that, at midday in direct sunlight, the illumination at the river substrate (15 cm depth) was 42% of surface photosynthetically active radiation (PAR) at the "light" station, and 0.3% of surface PAR under the sun shade ("dark" station).

The bricks were placed in the river on 24 July and recovered on 25 August 1977. Some of the dry bricks were immersed in synthetic crude oil (Table 20) for one minute on 24 July and then positioned in the river. The bricks were arranged in rows so that those which were unoiled would not be contaminated by oil.

The order of removal of each row of bricks was from downstream moving progressively upstream so as to minimize disturbances to the macrobenthos. Each brick was quickly lifted back into a nitex net (202 µm aperture) which was held just downstream. Each brick was thoroughly scrubbed with small brushes and the organisms which were recovered were immediately preserved in 10% formalin. Ten oiled and unoiled bricks from each station were used for macrobenthic analyses. The organisms were picked from debris in the laboratory under 10X magnification and all, except the Nematoda, Acari, and early instar Chironomidae, were identified and counted at a generic or specific level. The means from the 10 replicate samples were tested for significance with a 2 sample t-test after a log transformation of the original numbers (Elliott 1971).

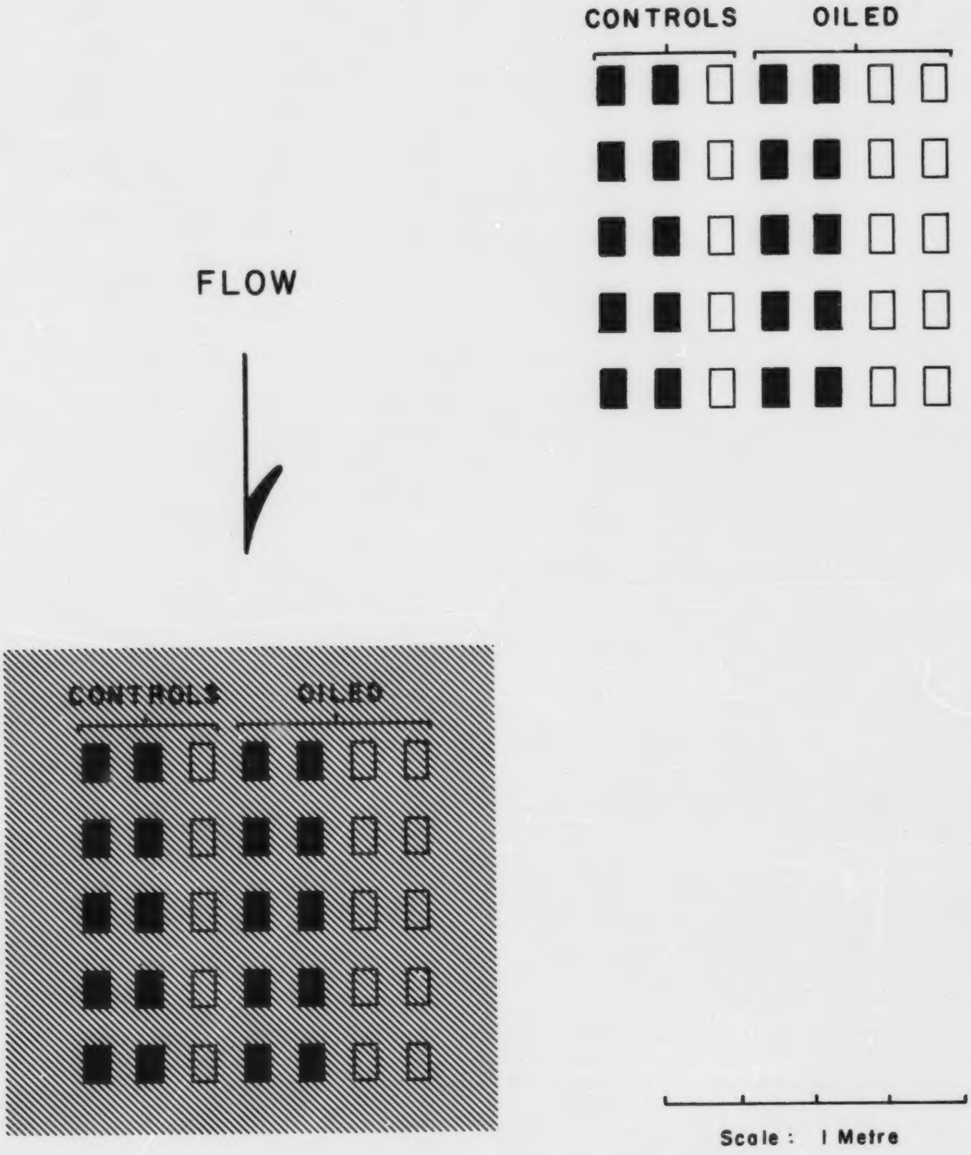

Figure 30. Experimental plan for the stations of bricks in the light (upper) and the dark (lower). (Dark bricks--macrobenthic samples; light bricks--microbenthic samples.)

Table 20. Physical-chemical characteristics of synthetic crude oil.

Characteristic	Value
Specific Gravity ($60^{o}/65^{o}$F)	0.848
Pour Point	5^{o}F
A.P.I.	35.4
Viscosity	
Absolute Centipoises	
35^{o}F	6.54
50^{o}F	4.30
70^{o}F	3.09
Kinematic Centistokes	
30^{o}F	7.61
50^{o}F	5.05
70^{o}F	3.66
Total Phenolics	0.45 $mg \cdot L^{-1}$
Total Sulphur	1550 $mg \cdot L^{-1}$
Predominant Hydrocarbon Types (% Volumes)	
Saturates	74.8
Aromatics	24.2
Olefins	1.0

Microbial biomass determinations were carried out on five separate bricks from each of the four treatments. Enumeration of sessile bacteria, based upon the direct counting method of Geesey et al. (in prep.), involved removing a 4 cm^2 area of epilithon using a template and sterile scalpel, and fixing this in cacodylate buffered (0.067M, pH 8.0) 0.5% glutaraldehyde. These samples were blended, appropriately diluted, stained with acridine orange, and the bacteria counted using epi-fluoresence microscopy. A log transformation was carried out on the bacterial counts (Elliott 1971) to obtain the geometric mean and 95% confidence limits. ATP biomass was determined on 4 cm^2 aseptic scrapes of epilithon using the technique of Holm-Hansen and Booth (1966). Chlorophyll *a* (an algal biomass indicator) was determined by scraping a 4 cm^2 area as above, transferring this to a vial containing filtered river water, and then filtering onto a Gelman glass fibre filter to which magnesium carbonate was added prior to freezing. Pigments were later extracted and assayed using the method of Moss (1967a, 1967b). Four square centimetre scrapes were also taken of the epilithon for direct examination of algae. Each sample was preserved with Lugol's Iodine and algae were identified and counted using the Utermohl sedimentation technique. The counts were transformed (log) to give the geometric mean and 95% confidence limits (Elliott 1971).

4.1.3 Results

On comparing the means of the number of bacteria·cm^{-2} on the control and oiled bricks in the light (Figure 31, Table 21), a highly significant difference was found between them ($p < 0.001$); a 9-fold increase occurred on the oiled bricks as opposed to the controls (Table 22). Similarly, in the dark (Table 23), there was a 4.5-fold increase in the number of bacteria on oiled bricks as opposed to the controls ($p < 0.002$). A comparison of the numbers of bacteria on the control bricks in the light and dark revealed slightly more bacteria present in the dark, but this difference was not significant. The same comparison for the oiled bricks revealed that

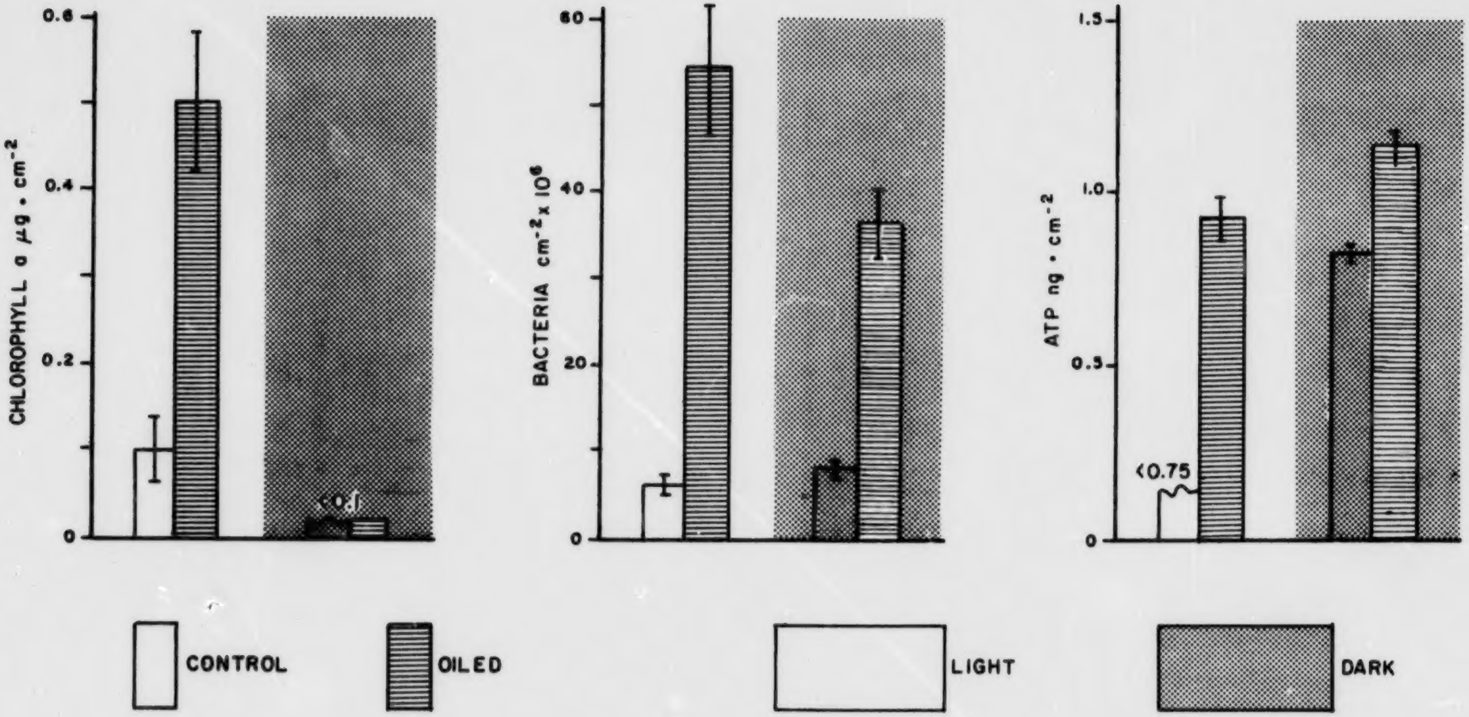

Figure 31. Counts of bacteria (mean with 95% confidence limits), chlorophyll *a* (mean ± SE), and ATP (mean ± SE) from oiled and unoiled bricks in the light and dark, as shown.

Table 21. Microbial biomass in the light on control and oiled limestone bricks. Bacteria, Cyanophyta, and Bacillariophyta, mean ± 95% confidence limits; chlorophyll *a* and ATP, mean ± SE.

Organism	Light - Control	Light - Oiled
Bacteria ($\text{Number}\cdot\text{cm}^{-2}$)	$6.0 \pm 0.6 \times 10^6$ ($n = 5$)	$5.4 \pm 0.6 \times 10^7$ ($n = 5$)
Cyanophyta ($\text{Number}\cdot\text{cm}^{-2}$)	$1.0 \pm 0.1 \times 10^5$ ($n = 4$)	$1.7 \pm 0.2 \times 10^5$ ($n = 4$)
Bacillariophyta ($\text{Number}\cdot\text{cm}^{-2}$)	$3.0 \pm 0.5 \times 10$ ($n = 4$)	$1.7 \pm 0.2 \times 10$ ($n = 4$)
Chlorophyll *a* ($\mu\text{g}\cdot\text{cm}^{-2}$)	0.1 ± 0.1 ($n = 4$)	0.5 ± 0.1 ($n = 5$)
ATP ($\text{ng}\cdot\text{cm}^{-2}$)	<0.75 ($n = 5$)	0.9 ± 0.1 ($n = 5$)

Table 22. A comparison of the means of the transformed (log) numbers of bacteria, Cyanophyta, and Bacillariophyta, and the arithmetic means of the levels of chlorophyll *a* and ATP using Student's t test.

ORGANISM	LIGHT - CONTROL LIGHT - OILED		DARK - CONTROL DARK - OILED		LIGHT - CONTROL DARK - CONTROL		LIGHT - OILED DARK - OILED	
	t	df	t	df	t	df	t	df
Bacteria	14.57^c	8	4.99^b	8	3.58^b	8	5.01^b	8
Cyanophyta	3.64^a	6	2.04^{NS}	8	1.42^{NS}	7	2.03^{NS}	7
Bacillariophyta	8.90^c	6	0.05^{NS}	8	5.62^c	7	6.45^c	7
Chlorophyll *a*	3.87	7	N/A		N/A		N/A	
ATP	N/A^d		3.21^a	8	N/A		2.43^a	8

^{a} $p<0.05$

^{b} $p<0.01$

^{c} $p<0.001$

NS - Not significant

N/A - Not applicable

Table 23. Microbial biomass in the dark on control and oiled limestone bricks.

ORGANISM[a]	DARK - CONTROL	DARK - OILED
Bacteria (Number·cm^{-2})	$8.0 \pm 1.0 \times 10^6$	$3.6 \pm 0.3 \times 10^7$
Cyanophyta (Number·cm^{-2})	$7.5 \pm 1.2 \times 10^4$	$1.2 \pm 0.2 \times 10^5$
Bacillariophyta (Number·cm^{-2})	$7.9 \pm 1.3 \times 10^2$	$8.1 \pm 0.3 \times 10^2$
Chlorophyll *a* (µg·cm^{-2})	<0.1	<0.1
ATP (ng·cm^{-2})	0.87 ± 0.06	1.24 ± 0.09

[a] The bacterial and algal counts had to be transformed (Elliott 1971) which allowed the 95% confidence limits to be calculated. For Chlorophyll and ATP, as n<30, only the S.E. could be computed (Elliott 1971).

significantly more bacteria were present in the light ($p \leq 0.05$), but this represented only a 50% increase over the numbers occurring in the dark.

Considerably more chlorophyll *a* was found to be present on the oiled bricks in the light as opposed to the control bricks in the light ($p \leq 0.01$), amounting to a 5-fold increase (Table 21). However, the levels of chlorophyll *a* on both oiled and control bricks in the dark was below the detection limit of the technique used (i.e., <0.1 µg chlorophyll $a \cdot cm^{-2}$) (Table 23). However, a direct count of algae revealed the presence of a considerable number of Bacillariophyta and Cyanophyta in the dark as well as the light (Figure 32, Tables 21, 22 and 23). Under light conditions, there was a 5.64-fold increase in the number of Bacillariophyta on the oiled as opposed to the control bricks ($p < 0.001$). Species dominance also differed, with *Cocconeis placentula* dominant on the control bricks and *Gomphonema olivaceus* and *Achnanthes minutissima* co-dominant on the oiled bricks. The numbers of Cyanophyta differed between the oiled and control bricks in the light; a 72% increase occurred on the oiled bricks in comparison with the controls ($p < 0.05$). No species composition differences were evident between the oiled and control bricks, with *Phormidium tenue* and *Lyngbya* spp.) particularly *L. servgineo-caerula,* being co-dominant in each case.

There was no significant difference between the numbers of Bacillariophyta or Cyanophyta occurring in the dark on oiled and control bricks. In terms of species composition, *Cocconeis placentula* and *Achnanthes minutissima* were the co-dominant Bacillariophycae, and *Phormidium tenue* and *Lyngbya* spp. were the co-dominant Cyanophycea algae. However, a third cyanophycean, *Schizothrix tinctoria,* was present in the dark in substantial numbers (<19% of the total) on both oiled and control bricks.

A comparison of the controls in the light and dark revealed a considerably greater number of bacillariophycean algae occurring in the light, $3.0 \pm 0.4 \times 10^3$ and $7.9 \pm 1.4 \times 10^2 \cdot cm^{-2}$, respectively,

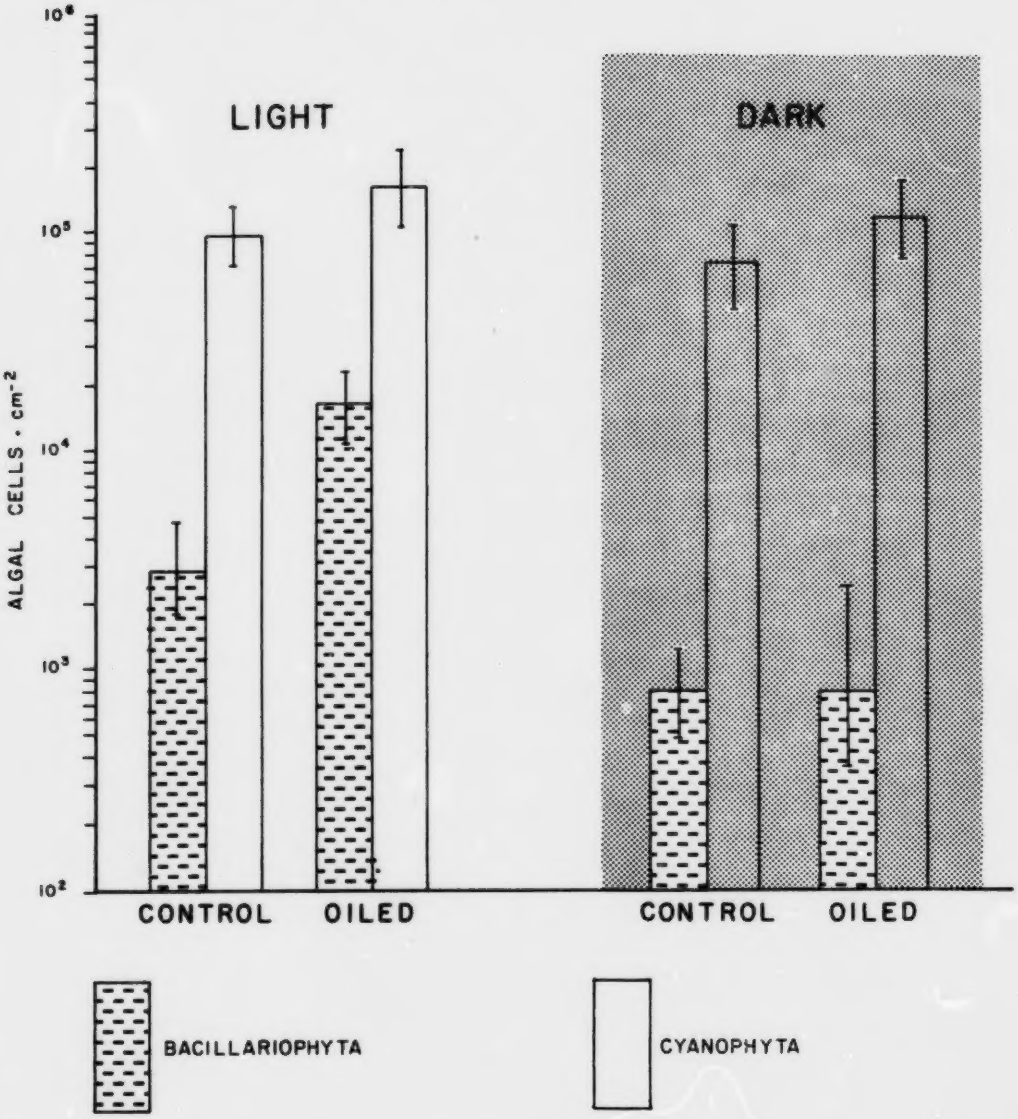

Figure 32. Algal cells·cm^{-2} (mean with 95% confidence limits) on oiled and unoiled bricks in the light and dark, as shown.

a highly significant 3.8-fold increase ($p \leq 0.001$). The same comparison for the cyanophycean algae gave $1.0 \pm 0.1 \times 10^5 \cdot cm^{-2}$ in the light and $7.5 \pm 1.4 \times 10^4 \cdot cm^{-2}$ in the dark, and the difference between them is not significant. On comparing the numbers of bacillariophycean algae on the oiled bricks, in the light there were $1.7 \pm 0.2 \times 10^4 \cdot cm^{-2}$ and in the dark $8.1 \pm 2.7 \times 10^{-2}$, a 21-fold increase ($p \leq 0.001$). However, there was no significant difference between the numbers of cyanophycean algae present on the oiled bricks in the light or dark, $1.7 \pm 0.1 \times 10^5 \cdot cm^{-2}$ and $1.2 \pm 0.2 \times 10^5 \cdot cm^{-2}$, respectively. A complete list of algal species is shown in Appendix 6.2.

The levels of ATP (Figure 31, Tables 21, 22, and 23) on all bricks were very similar (≈ 1 ng $ATP \cdot cm^{-2}$). The only significant difference was between the oiled and control bricks in the dark, with 42% more occurring on the oiled substrate ($p \leq 0.01$).

A comparison of the mean numbers of macrobenthos on bricks in the light revealed that there were significantly fewer Acari and Hydropsychidae on the oiled than on the control bricks (Table 24). There were significantly more *Lepidostoma*, Chironomidae (in particular, the Orthocladiinae and Tanypodinae), Naididae, and total numbers of taxa on the oiled bricks as compared with the controls. In the dark, there were significantly fewer Gastropoda and *Baetis* sp. on the oiled bricks than on control ones and significantly more Tanypodinae (Table 24).

When the control (unoiled) bricks were compared in the light and the dark, the only significant differences were for *Baetis* sp. (more in the dark) and Hydropsychidae (less in the dark) (Table 25). A comparison of the oiled bricks in the light and in the dark revealed that there were significantly fewer Sphaeriidae, *Lepidostoma* sp., Chironomidae (particularly Orthocladiinae), Naididae, and total benthos on the control bricks in the light, but significantly more Heptageniidae (Table 25). A summary of the oil-sun-light interactions for macrobenthos is given in Table 26.

Table 24. Comparison of the geometric means of the numbers of macrobenthos for 10 replicates. Control (unoiled) bricks vs. oiled bricks in direct sunlight and in the shade.

Organism	Light			Dark		
	Control	Oiled	t	Control	Oiled	t
Acari	2.2	0.9	2.22[a]	2.3	0.8	2.03
Sphaeriidae	2.4	3.5	0.60	1.3	0.8	0.62
Gastropoda	1.7	0.8	1.44	2.8	0.5	2.40[a]
Ephemeroptera	4.4	3.0	1.03	6.4	4.6	1.02
Baetis sp.	1.6	0.9	1.00	4.5	0.9	4.34[b]
Heptageniidae	2.3	1.3	1.30	1.9	3.5	1.81
Plecoptera	1.9	0.8	1.62	2.5	1.1	1.55
Trichoptera	9.8	9.6	0.84	4.9	7.4	1.07
Ceraclea sp.	3.7	2.9	0.51	1.9	2.5	0.50
Lepidostoma sp.	1.1	4.0	3.48[b]	0.8	1.1	0.67
Hydropsychidae	3.1	0.8	2.84[b]	1.2	2.2	1.20
Chironomidae	4.0	14.9	4.02[b]	5.4	6.9	0.71
Orthocladiinae	2.9	10.8	4.23[b]	3.6	4.7	0.75
Tanytarsini	0.8	1.4	0.82	1.3	1.2	0.11
Tanypodinae	0.1	1.4	4.49[b]	0.0	0.6	2.65[b]
Naididae	0.0	2.7	5.71[b]	0.2	0.2	0.06
Total Taxa	13.2	17.0	2.20[a]	11.4	10.0	0.38
Total Benthos	27.8	43.4	2.03	24.7	23.8	0.12

[a] $p < 0.05$

[b] $p < 0.01$

Table 25. Comparison of the geometric means of the numbers of macrobenthos for 10 replicates. Control (unoiled) bricks in light and dark and of oiled bricks in the light and dark.

Organism	Control			Oiled		
	Light	Dark	t	Light	Dark	t
Acari	2.2	2.3	0.02	0.9	0.8	0.24
Sphaeriidae	2.4	1.3	0.85	3.5	0.8	2.20[a]
Gastropoda	1.7	2.8	0.80	0.8	0.5	0.63
Ephemeroptera	4.4	6.4	1.20	3.1	4.5	1.08
Baetis sp.	1.7	4.5	2.37[a]	1.5	0.9	1.01
Heptageniidae	2.3	1.9	0.60	1.3	3.5	2.39[a]
Plecoptera	1.9	2.5	0.52	0.7	1.1	0.73
Trichoptera	9.8	4.9	1.97	9.6	7.4	1.03
Ceraclea sp.	3.7	1.9	1.28	3.0	2.5	0.32
Lepidostoma sp.	1.1	0.8	0.73	4.0	1.1	3.48[b]
Hydropsychidae	3.1	1.2	2.11[a]	0.8	2.2	1.90
Chironomidae	4.0	5.4	0.75	14.7	6.7	2.80[a]
Orthcladiinae	2.9	3.6	0.65	10.8	5.4	2.74[a]
Tanytarsini	0.7	1.3	0.79	1.4	1.2	0.35
Tanypodinae	0.07	0	1.0	1.4	0.6	1.64
Naididae	0.0	0.25	1.5	2.7	0.2	4.03[b]
Total Taxa	13.2	11.4	0.81	17.1	10.0	1.68
Total Benthos	27.8	24.7	0.39	43.3	23.8	2.49[a]

[a] $p < 0.05$

[b] $p < 0.01$

Table 26. The response of macrobenthos to oil-light or dark conditions on bricks.

Organism	Dark - Oiled Bricks vs. Unoiled Control	Light - Oiled Bricks vs. Unoiled Control
Acari	0[a]	-
Gastropoda	-[b]	0
Ephemeroptera	0	0
Baetis sp.	-	0
Heptageniidae	0	0
Trichoptera		
Lepidostoma sp.	0	+
Hydropsychidae	0	-
Chironomidae	0	+
Orthocladiinae	0	+
Tanypodinae	+[c]	+
Naididae	0	+
Total Taxa	0	+
Total Benthos	0	0

[a] no significant difference between controls

[b] significant reduction

[c] significant increase in mean numbers

4.1.4 Discussion

The attempt to examine the community response of the micro- and macrobenthic organisms in the presence and absence of algae indicated that, even though the level of light had been reduced to 0.3% of the surface photosynthetically active radiation (PAR), algal populations were still present underneath the shade. Evidence that this was not an experimental artifact came from a concurrent study (Lock et al. in prep.) on the bacteria and algae inhabiting the lower surfaces of rocks where the PAR was 0.03% of that at the surface. In the latter study, substantial populations of cyanophycean and bacillariophycean algae were found on the lower surfaces of granite discs and epoxy plates using light microscopy and scanning electron microscopy.

In both the light and dark, there was a considerable increase in the numbers of bacteria present on the oiled bricks as opposed to the unoiled control, 9-fold and 4.5-fold, respectively. Such a response had been noted in an experimental addition of crude oils to soils (Sexstone and Atlas 1977), an Arctic lake (Hutchinson et al. 1976), the Beaufort Sea area (Atlas 1977), and artificial ponds (Shindler et al. 1975). It seems reasonable to assume that at least a portion of this increase was due to a proliferation of oil degrading bacteria (Walker and Colwell 1977). This conclusion is supported below (Section 4.3) where Muskeg River sediments were demonstrated to have a substantial ability to degrade the saturate fraction of synthetic crude oil.

The responses of algae to crude oil are extremely varied, ranging from stimulation through to tolerance or inhibition, dependent upon algal species, the length of incubation, and the composition and initial concentration of the oil tested (Kauss et al. 1973; Pulich et al. 1974; Kauss and Hutchinson 1975; Graham and Hutchinson 1975; Parsons et al. 1976). In this study, an overall stimulation was found in algal biomass over that occurring on unoiled bricks under light conditions. This was apparent through direct counts and by chlorophyll *a* determinations. The greatest change was in the Bacillariophyta, which showed a 5.6-fold increase in numbers in the presence of oil with a concomitant shift in species composition. *Cocconeis placentula* was dominant in the control and *Gomphonema olivaceum* and *Achnanthes minutissima* were co-dominant on the oiled bricks. Numbers of Cyanophyta were also significantly different between the oiled bricks and the control with a 72% increase on the oiled; however, no major shifts in species composition were evident. In the dark, there was no significant difference between the numbers of Bacillariophyta or Cyanophyta occurring on oiled or control bricks, nor were there any shifts in species composition. It would, therefore, seem that the stimulatory action of synthetic crude oil upon algae is not evident under conditions of very low light (<0.3% of surface PAR).

A comparison of the algal standing crop in the light and dark revealed a very much greater proportion (3.8-fold increase for control bricks and a 21-fold increase for oiled bricks) of Bacillariophyta occurring in the light. But, the same comparison for the Cyanophyta revealed that there is not significant difference in the numbers occurring in the light and dark on oiled or control bricks. This finding contrasts with those of Roeder et al. (1975) working on lakes and rivers in the Northwest Territories, where they found apparent enhancement of cyanophycean algae by Norman Wells crude oil.

On oiled substrates in the light, there were significantly more taxa (*Lepidostoma*, Chironomidae, Orthocladiinae, Tanypodinae, Naididae, and total taxa) than on the unoiled control bricks (Tables 25 and 26). This positive response may be due either to an increase in the standing crop of microbes or algae for macrobenthic grazers, or to an improved structure of the substrate, caused by oil-microbial interactions, as discussed below. Fewer Acari and Hydropsychidae were found on oiled, sunlit bricks. In the case of the Hydropsychidae, this may be a response to the greater growth of the microbial film on the oily substrate which may have actively interfered with the nets constructed by the animals. On oiled substrates in the dark, there were significantly more Tanypodinae, but fewer *Baetis* and Gastropoda (Table 24). This may be a consequence of the characteristic preference of many benthic animals for sunlit or dark rock surfaces (Hynes 1970).

On unoiled substrates, significantly fewer *Baetis* were found in the light, but more Hydropsychidae (Table 24). Similarly, for the oiled substrates, more *Lepidostoma,* Orthocladiinae, Naididae, and total benthos were found in the light, but fewer Heptageniidae. As previously noted, the lower numbers on the dark bricks may be a reflection of the preference of some macrobenthos for upper rock surfaces. The preference of many organisms for the oiled bricks in the light is probably a direct response to the increased bacterial and algal biomass (i.e., food) occurring on these bricks.

4.2 THE EFFECTS OF SYNTHETIC CRUDE OIL AND ITS COMPONENT OILS ON ESTABLISHED BENTHIC COMMUNITIES

4.2.1 Introduction

This study examined the effects of synthetic crude oil and its component fractions (naphtha, gas-oil, and kerosene) on established microbial and macrobenthic communities on artificial, limestone substrates in a brown-water river. As such, this experimental treatment simulated a large spillage of refined oils into a characteristic brown-water river of the Athabasca Oil Sands region.

Parker et al. (1976) noted that most studies on hydrocarbon toxicity deal with the short-term effects (bioassays), but that the longer term effects of sublethal dosages of such chemicals represented a gap in our knowledge which warranted priority attention. Light refined hydrocarbons are known to exert an immediate, toxic effect on aquatic biota (Berry and Brammer 1977). Other studies (Rosenberg and Wiens 1976) have studied the effects of oils on dry, artificial substrates, which were subsequently immersed in rivers. However, in the present study, the interest was in the length of time and the nature of the recovery of established benthic communities from acute spillages. Further, since component fractions of synthetic crude oil were available, it was possible to critically assess the relative toxicities and persistence of each type of oil. By so doing, it was hoped to be able to obtain new insights into the relative importance of such oil fractions in determining the toxicity of synthetic oils on freshwater biota.

4.2.2 Materials and Methods

This experiment was done in the Muskeg River about 500 m downstream from the light-dark oiled brick experiment (Section 4.1). Five stations of bricks were set up in a uniform riffle area, each station being about 2 m downstream from the next (Figure 33). The uppermost station consisted of 12 bricks laid across the river, each parallel to the flow, and the lower four stations were composed of 46 bricks each. The stations were arranged on a square matrix pattern. A block of five rows and five columns of bricks at each station were used for the macrobenthic experiment (five replicates for each of the four oil types and the control). A similar matrix (3 x 7) containing the remaining 21 bricks were used for the microbial biomass determinations. A non-randomized pattern was used to minimize the possibility of contamination of downstream bricks by different oils and to simplify the recovery of bricks in the winter through ice. Each column of each oil type was positioned to minimize any contamination from upstream bricks by differing oils, although what little oil did erode from the bricks appeared to rise quickly to the water surface.

Dry bricks were placed in the river on 17 May and about six weeks later, on 28 June 1977, the pre-treatment, control station was sampled to characterize pre-treatment communities. The remaining appropriate bricks were immersed in the oils (Figure 33).

The bricks were lifted from the substrate and completely immersed for 1 min in the appropriate oil (naphtha, kerosene, gas-oil, or synthetic crude oil). The control bricks were lifted and dipped into clean pails of water. The bricks were then replaced

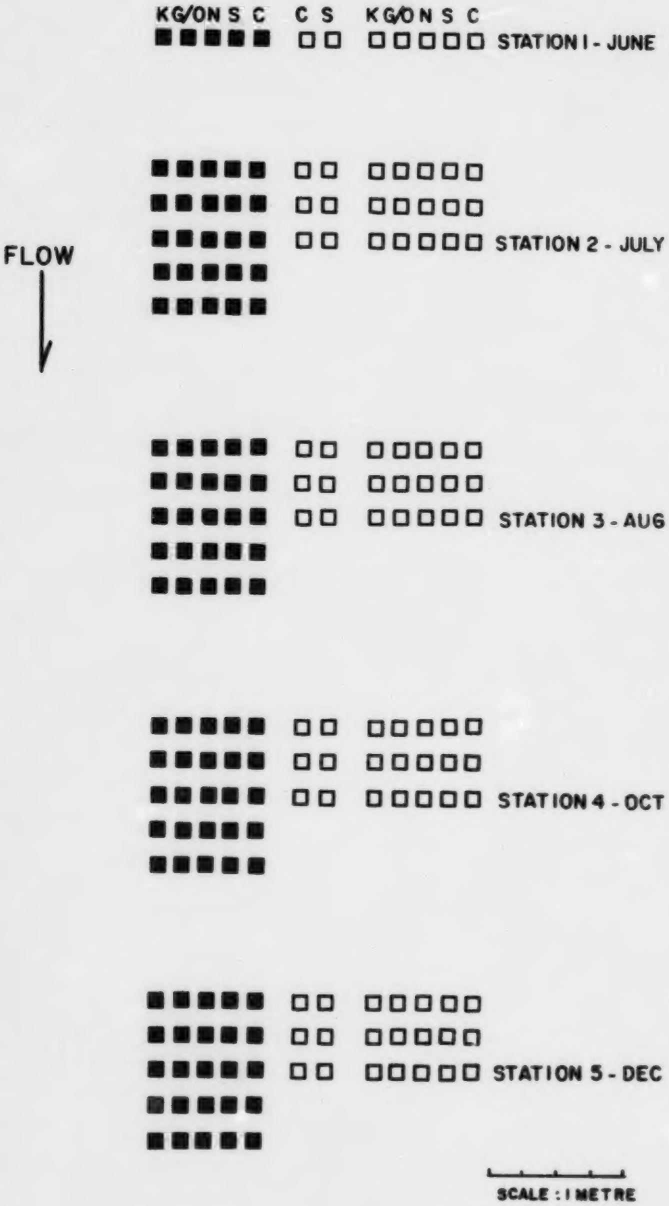

Figure 33. Schematic diagram of the stations established for contamination studies with synthetic crude oil (S) and component fractions, kerosene (K), gas-oil (G/O), naphtha (N), and the unoiled control (C). The dark and light-colored bricks were those used for macro- and microbenthic analyses, respectively.

in their former positions. At the time that the bricks were oiled, they were coated with a substantial algal and bacterial growth which caused the oil to form a bright sheen over the bricks upon immersion.

Stations 2 through to 5 were recovered on 26 July, 23 August, 26 October, and 6 December, respectively.

Each brick for macrobenthic analyses was recovered by quickly lifting it from the substrate into a 202 μm Nitex net held just downstream from it. Each station was removed by working from downstream to minimize disturbances on nearby bricks.

The bricks, once recovered, were taken to shore and scrubbed with small brushes. Macroinvertebrates were concentrated with a 180 μm sieve and were immediately preserved in 10% formalin. In the laboratory, the organisms were washed, picked out from the debris under 10X magnification, enumerated and identified as to species or genus, and preserved in 70% ethanol.

The only deviation from this procedure occurred for Station 5 which was recovered on 6 December at a temperature of -45°C. A hole was cut in the 25 cm thick ice around the brick station and the bricks were lifted from the river directly into aluminum foil wrappers, all of which froze almost immediately. Sampling nets could not be used as they froze immediately. The samples were kept frozen until they were analyzed in the laboratory, at which time they were thawed and preserved in 70% ethanol.

Statistical tests (F-test and Student's t) were done on five replicate samples with a 2 sample t test after a log transformation (Elliott 1971) for genera, orders, total taxa, and numbers of species for the July and August dates (Table 8).

Microbial samples collected in December were prevented from freezing by maintaining them in a large container of water which was kept from freezing by the additions of small amounts of warm, sterile water until the samples could be air-lifted back to the Mildred Lake laboratory. Microbial biomass determinations were carried out on 4 cm scrapes of epilithon as described previously (Section 4.1). Bacteria were enumerated by direct counting using acridine orange staining and an epifluorescent microscope and algae by the Utermohl sedimentation technique. Chlorophyll *a* was determined using the method of Moss (1967a, 1967b) and ATP by the method of Holm-Hansen and Booth (1966).

4.2.3 Results

Densities of macroinvertebrates, bacteria, and algae on the bricks all varied between sampling dates in a similar pattern (Figures 34, 35, 36, and 37). The total numbers of macrobenthos for each treatment remained similar throughout the study. After a decrease in total numbers in August, there was a substantial increase in October and December which was correlated with bacterial/algal counts. There were few significant differences between individual taxa or groups of benthos between the controls and the oiled bricks at each of the stations throughout the experiment (Tables 27 and 28).

The Chironomidae constituted the largest proportion of the total numbers (Table 28). In July, the Trichoptera, as a group, were significantly ($p \leq 0.05$) less abundant on the synthetic crude oil-contaminated bricks as compared with the controls, although none of the values for individual genera were significant (Table 27). *Baetis* was significantly ($p \leq 0.01$) less abundant on the gas-oil and kerosene-contaminated bricks, whereas *Glossosoma* and *Corynoneura* were significantly ($p \leq 0.01$) more abundant on the kerosene-contaminated bricks, although both occurred only in small numbers. *Simulium* were significantly ($p \leq 0.05$) less abundant on the naphtha- and kerosene-treated bricks.

In August, *Heptagenia* and *Cheumatopsyche* were significantly ($p \leq 0.05$) less abundant on the gas-oil- and synthetic crude oil-contaminated bricks, respectively (Table 27). The Acari and *Eukiefferiella* were significantly more abundant ($p \leq 0.05$) on the kerosene-treated bricks. Total Chironomidae and the total numbers of benthos were significantly ($p \leq 0.05$) greater than control levels (Table 28) on the naphtha-treated bricks. No significant differences were detected for the groups in October or December.

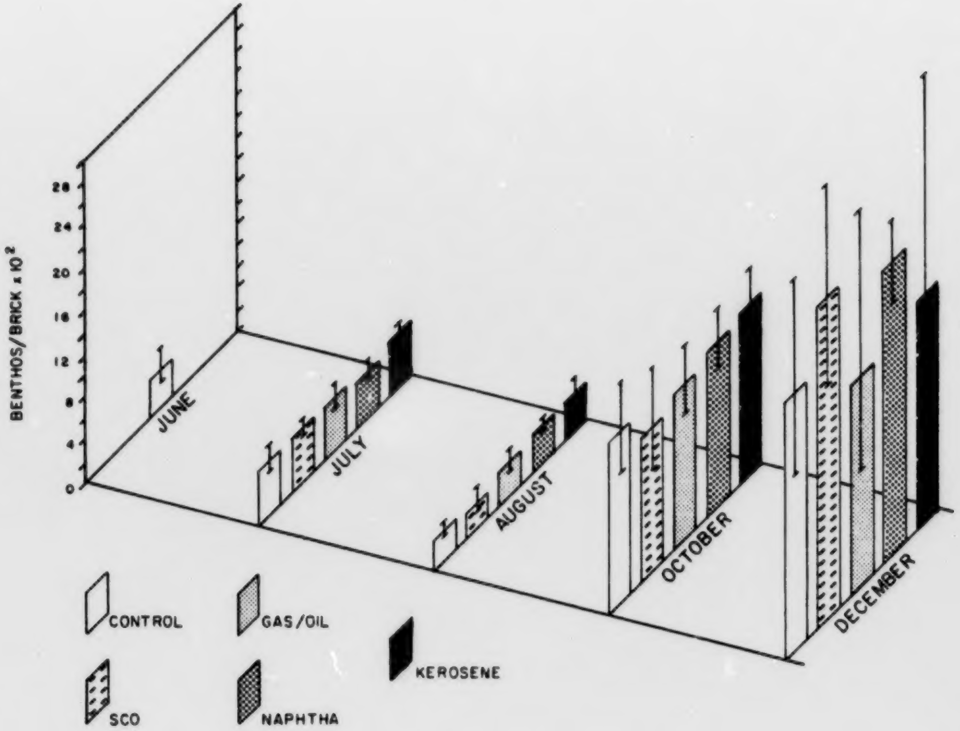

Figure 34. Total numbers of macrobenthos per brick. There were five replicates for each sample; the bars show the means with 95% confidence limits (SCO--synthetic crude oil).

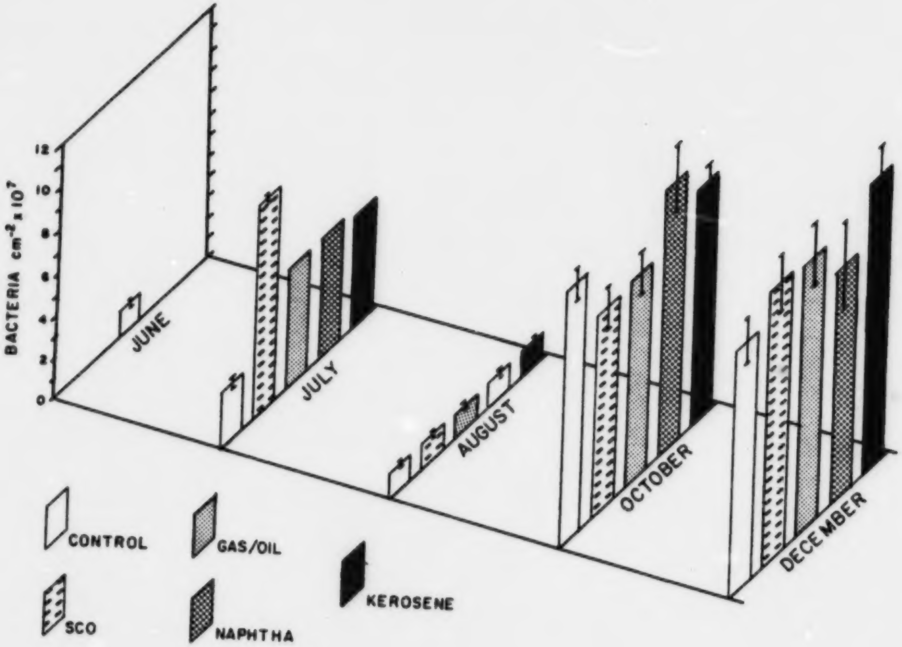

Figure 35. Mean bacterial counts cm^{-2} x 10 based on five replicate samples with 95% confidence limits (except for July gas-oil, naphtha, and kerosene which were one-brick samples) (SCO--synthetic crude oil).

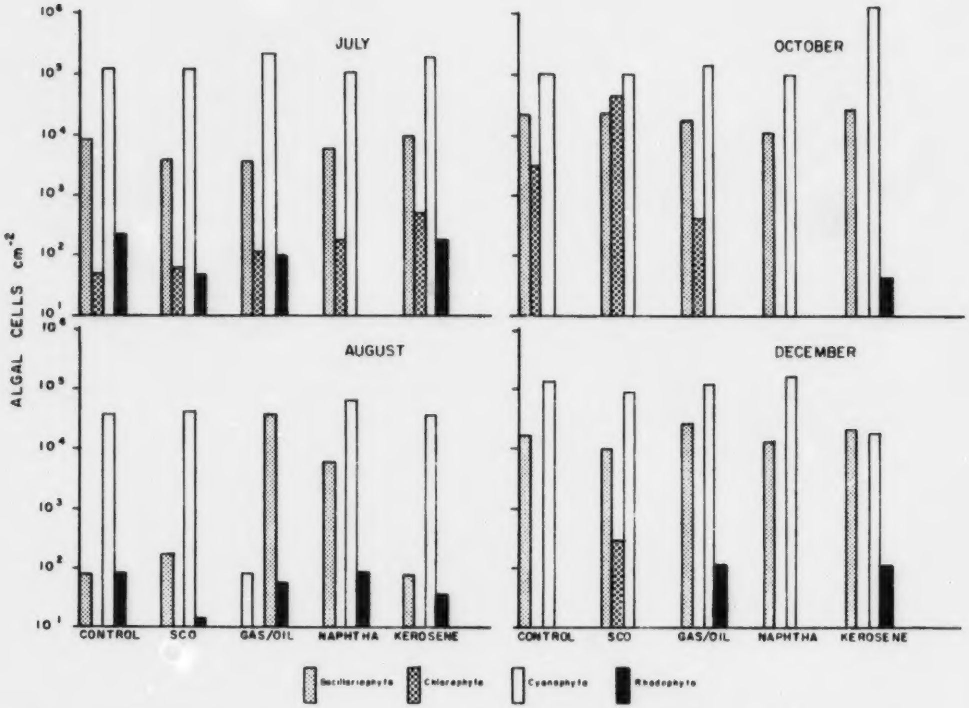

Figure 36. Numbers of algal cell·cm^{-2} on the control and oil-coated bricks (SCO--synthetic crude oil).

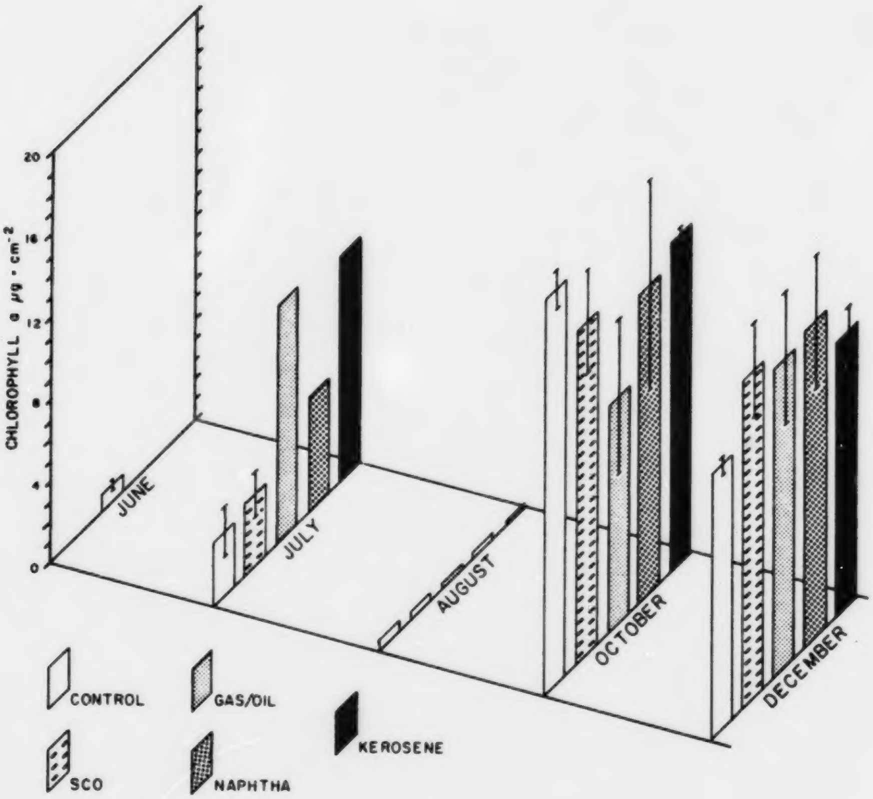

Figure 37. Mean chlorophyll *a* on five replicate samples with 95% confidence limits (except for July gas-oil, naphtha, and kerosene which were one-brick samples) (SCO--synthetic crude oil).

Table 27. Total numbers of organisms per brick as a geometric mean (5 replicates)^{a}.

DATE	TREATMENT^{c}	ORGANISM							
		Heptagenia	*Baetis*	*Pseudocleon*	*Hydropsyche*	*Psychomyia*	*Cheumatopsyche*	*Glossosoma*	Acari
26 July	Control	6	27	42	27	8	4	3	5
	SCO	5	22	42	13	6	2	2	7
	G/O	4	13^{b}	41	30	9	2	4	6
	Naph	4	19	39	23	12	2	2	6
	Ker	6	17^{b}	36	27	14	5	7^{b}	7
23 Aug	Control	3	9	1	8	9	8	1	9
	SCO	2	8	0	5	11	2^{a}	1	12
	G/O	2^{a}	10	1	9	8	2^{a}	2	9
	Naph	3	8	1	12	10	4	1	15
	Ker	3	5	1	14	8	5	1	17^{a}

DATE	TREATMENT	*Corynoneura*	*Eukieffriella*	*C. bicinctus*	*Synorthocladius*	*Orthocladiinae*	*Rheotanytarsus*	*Simulium*	Total Species
26 July	Control	2	16	25	49	242	8	85	30
	SCO	2	13	31	51	271	7	28	29
	G/O	2	16	33	39	273	5	47	29
	Naph	2	15	36	47	245	7	21^{a}	28
	Ker	4^{b}	11	43	55	297	5	27^{a}	31
23 Aug	Control	3	6	37	9	5	8	-	31
	SCO	2	4	22	10	3	9	-	32
	G/O	5	9	13	7	5	17	-	33
	Naph	3	10	47	11	5	13	-	38
	Ker	4	19^{a}	50	14	4	16	-	35

[a] $p < 0.05$

[b] $p < 0.01$

[c] Symbols; SCO - Synthetic crude oil, G/O - Gas-oil, Naph - Naphtha, Ker - Kerosene.

Table 28. Total numbers of organisms per brick as a geometric mean (5 replicates).

Date	Treatment[b]	Organism			
		Chironomidae	Ephemeroptera	Trichoptera	Total Benthos
26 July	Control	242	79	53	518
	SCO	271	73	34[a]	486
	G/O	273	61	57	500
	Naph	245	66	51	427
	Ker	299	71	44	506
23 Aug	Control	192	22	41	284
	SCO	156	17	31	237
	G/O	213	22	41	318
	Naph	243[a]	27	45	354[a]
	Ker	248	18	42	348
26 Oct	Control	1415	72	35	1574
	SCO	1186	49	38	1324
	G/O	1304	54	28	1446
	Naph	1359	60	34	1516
	Ker	1384	58	39	1565
6 Dec	Control	2266	19	20	2332
	SCO	2834	19	19	2899
	G/O	1842	7	17	1889
	Naph	2537	16	23	2604
	Ker	1988	11	23	2041

[a] $p < 0.05$

[b] Symbols: SCO - Synthetic crude oil
G/O - Gas-oil
Naph - Naphtha
Ker - Kerosene

After colonizing for six weeks, the numbers of bacteria$\cdot$cm^{-2} had reached 1.3 ± 0.1 x 10^7 (Figure 35, Table 29). One month after the application of the oils, the number of bacteria on the control had reached $2.7 \pm 0.3 \times 10^7 \cdot cm^{-2}$. However, by July, the numbers present on the bricks which had received oil were very much higher, ranging from 3.7-fold more on the synthetic crude oil, to about 2-fold less for gas-oil, naphtha, and kerosene, the numbers$\cdot$cm^{-2} being $1.0 \pm 0.2 \times 10^8$ on the former and 5.2×10^7 to 5.6×10^7 for the other three components, respectively (Table 29, Figure 35). The difference between the numbers of bacteria$\cdot$cm^{-2} on the control and the number on the synthetic crude oil-treated brick (Table 30) was significant ($p < 0.001$). After July, there ceased to be any significant difference between the control and the synthetic crude oil or its three major components, except on synthetic crude oil- and gas-oil-treated bricks in October, when there were significantly ($p < 0.01$) less numbers of bacteria$\cdot$cm^{-2}. However, these reductions amounted to only 22% of the control (Table 29). During August, there were less total numbers of bacteria to around $1 \times 10^{-7} \cdot cm^{-2}$ for the oil-treated bricks, but the numbers for both control and oiled bricks increased in October and December (Figure 35).

On 28 June (pre-treatment), the algae present on the control bricks were dominated almost entirely by Cyanophyta with a few Bacillariophyta present (Table 31). One month after the oil additions, four major taxonomic groups were present: Bacillariophyta, Chlorophyta, Rhodophyta, and Cyanophyta. A significant reduction, compared with controls ($p < 0.001$), in the number of bacillariophycean algae was apparent on the synthetic crude oil- and gas-oil-treated bricks. Significant increases over controls of chlorophycean algae were noted on gas-oil, naphtha, and kerosene, ranging from 200 to 1000% ($p < 0.001$), and for Cyanophyta on gas-oil and kerosene of 145% and 175%, respectively ($p < 0.001$). The numbers of Rhodophyta were all significantly lower ($p < 0.05$ to 0.001) (Table 32).

Table 29. Numbers of bacteria·cm^{-2} x 10^7 on control and oiled limestone bricks over time, mean plus 95% confidence limits.

DATE	CONTROL	SYNTHETIC CRUDE OIL	GAS/OIL	NAPHTHA	KEROSENE
28 June (pre-treatment)	1.3±0.1 (n=5)	-	-	-	-
26 July	2.7±0.3 (n=4)	1.0±2.0 (n=4)	5.6 (n=1)	5.6 (n=1)	5.2 (n=1)
23 August	1.1±0.1 (n=6)	1.2±0.1 (n=6)	9.5±1.0 (n=3)	1.1±0.1 (n=3)	1.1±0.1 (n=3)
26 October	12.0±1.0 (n=6)	9.4±1.0 (n=6)	9.4±1.0 (n=3)	12.0±2.0 (n=3)	11.0±1.0 (n=3)
6 December	12.0±1.0 (n=6)	13.0±1.0 (n=6)	13.0±1.0 (n=3)	11.0±2.0 (n=3)	14.0±1.0 (n=3)

Table 30. Comparison of the means of the number of bacteria·cm^{-2} on the controls and synthetic crude oil and its three components.

Sample	July		August		October		December	
	t	df	t	df	t	df	t	df
Control vs. Synthetic Crude Oil	12.710^{a}	6	0.178$^{NS^{c}}$	10	3.711^{b}	10	1.664^{NS}	10
Control vs. Gas/Oil	N/A^{c}		1.805^{NS}	7	3.504^{b}	7	1.153^{NS}	7
Control vs. Naphtha	N/A		0.793^{NS}	7	0.629^{NS}	7	0.772^{NS}	7
Control vs. Kerosene	N/A		0.608^{NS}	7	1.601^{NS}	7	2.240^{NS}	7

[a] $p < 0.001$

[b] $p < 0.01$

[c] Symbols: N/A = Not Applicable
NS = Not Significant

Table 31. Bacillariophyta, Chlorophyta, Cyanophyta, and Rhodophyta in terms of numbers·cm^{-2} and expressed as a percent of the control for the four oils at the four sampling periods.

Date	Oil	Bacillariophyta		Chlorophyta		Cyanophyta		Rhodophyta	
		number·cm^{-2}	%	number·cm^{-2}	%	number·cm^{-2}	%	number·cm^{-2}	%
July									
	SCO	4.0×10^3	49	6.4×10	75	12.3×10^4	100	4.8×10	22
	Gas-oil	3.7×10^3	45	1.1×10^2	235	2.2×10^5	174	9.6×10	44
	Naphtha	6.1×10^3	75	1.8×10^2	368	1.1×10^5	92	-	0
	Kerosene	9.4×10^3	115	4.8×10^2	1000	1.8×10^5	145	1.8×10^2	80
	Control	8.1×10^3	-	4.8×10	-	12.4×10^4	-	2.2×10^2	-
August									
	SCO	1.8×10^2	221	1.0×10	-	4.2×10^4	109	1.4×10	17
	Gas-oil	8.2×10	100	-	-	3.8×10^4	98	5.6×10	68
	Naphtha	1.1×10^2	134	-	-	6.8×10^4	177	8.8×10	107
	Kerosene	7.6×10	95	-	-	3.5×10^4	91	3.8×10	46
	Control	8.0×10	-	-	-	3.9×10^4	-	8.2×10	-
October									
	SCO	2.2×10^4	106	4.3×10^4	1479	9.8×10^4	97	-	-
	Gas-oil	1.7×10^4	80	3.8×10^2	13	1.3×10^5	126	-	-
	Naphtha	9.9×10^3	48	-	0	8.9×10^4	88	-	-
	Kerosene	2.4×10^4	118	-	0	1.1×10^6	108	-	-
	Control	2.1×10^4	-	2.9×10^3	-	10.1×10^4	-	-	-

continued ...

Table 31. Concluded.

Date	Oil	Bacillariophyta		Chlorophyta		Cyanophyta		Rhodophyta	
		number·cm^{-2}	%	number·cm^{-2}	%	number·cm^{-2}	%	number·cm^{-2}	%
December									
	SCO	1.0×10^4	71	2.9×10^2	-	1.0×10^5	71	-	-
	Gas-oil	2.6×10^4	183	-	-	1.2×10^5	94	1.1×10^2	-
	Naphtha	1.3×10^4	89	-	-	1.6×10^5	125	-	-
	Kerosene	2.0×10^4	138	-	-	1.8×10^4	14	1.1×10^2	-
	Control	1.4×10^4	-	-	-	12.5×10^4	-	-	-

Table 32. A table of χ^2 values for the comparison of algal numbers on the control bricks with those on oiled bricks based upon the null hypothesis that no difference exists between them.

	Month			
	July	August	October	December
BACILLARIOPHYTA				
Synthetic crude oil	14.14[c]	36.60[c]	0.40^{NS}	7.22[b]
Gas-oil	16.76[c]	0.024^{NS}	4.32[a]	34.96[c]
Naphtha	2.89^{NS}[d]	3.63^{NS}	36.96[c]	0.83^{NS}
Kerosene	0.90^{NS}	0.10^{NS}	2.90^{NS}	8.87[b]
CHLOROPHYTA				
Synthetic crude oil	2.28	-	349.34[c]	-
Gas-oil	26.24[c]	-	19.36[c]	-
Naphtha	35.66[c]	-	-	-
Kerosene	353.40[c]	-	-	-
CYANOPHYTA				
Synthetic crude oil	0^{NS}	0.17^{NS}	0.03^{NS}	5.79[a]
Gas-oil	24.66[c]	0	3.09^{NS}	0.20^{NS}
Naphtha	0.45^{NS}	0.36[b]	0.76^{NS}	3.62^{NS}
Kerosene	10.47[c]	0.59^{NS}	0.28^{NS}	9.23[b]
RHODOPHYTA				
Synthetic crude oil	110.38[c]	48.16[c]	-	-
Gas-oil	48.64[c]	4.89[a]	-	-
Naphtha	220.00[c]	0.21^{NS}	-	-
Kerosene	4.65[a]	16.13[c]	-	-

[a] $p < 0.05$

[b] $p < 0.01$

[c] $p < 0.001$

[d] Symbol: NS: not significant

Two months after treatment, in August, the numbers of Bacillariophyta and Cyanophyta were not significantly different from the control except for the Bacillariophyta on the synthetic crude oil-treated bricks which were 100% higher ($p < 0.001$) and for the Cyanophyta on the naphtha-treated bricks which were 77% higher ($p < 0.01$). Numbers of rhodophycean algae were generally much lower than the control levels for bricks dipped in naphtha. Chlorophycean algae were absent from the control and all the oil-treated bricks, except for the synthetic crude oil, where $100 \cdot cm^{-2}$ were recorded.

In October, no significant differences were apparent between the numbers of Cyanophyta on the control and oiled bricks and for the bacillariophycean algae on synthetic crude oil- and kerosene-treated bricks. However, lower numbers were present on gas-oil and naphtha treatments ($p < 0.05$ and 0.001). The numbers of rhodophycean algae were very much higher on the synthetic crude oil-treated bricks, a 1479% increase ($p < 0.001$), but were reduced or close to zero on the other three oil treatments.

In December, the algae were primarily represented by Bacillariophyta and Cyanophyta. With the exception of Bacillariophyta on gas-oil and Cyanophyta on naphtha, the oiled treatments were not dissimilar from the controls (Figure 37). An algal species list is included in Appendix 6.2.

The level of chlorophyll $a \cdot cm^{-2}$ on the pre-treatment bricks was 1.3 ± 0.1 g in July (Figure 38, Table 33). One month after applying oil to the bricks, the level of chlorophyll *a* on the unoiled control was $3.15 \pm 1.2\ g \cdot cm^{-2}$, the level of synthetic crude oil was close to this at $3.46 \pm 1.12\ g \cdot cm^{-2}$, but the levels on gas-oil, naphtha, and kerosene were considerably higher, 11.58, 5.63, and $10.88\ g \cdot cm^{-2}$, respectively. Levels in August, October, and December were not significantly different from the controls (Table 34).

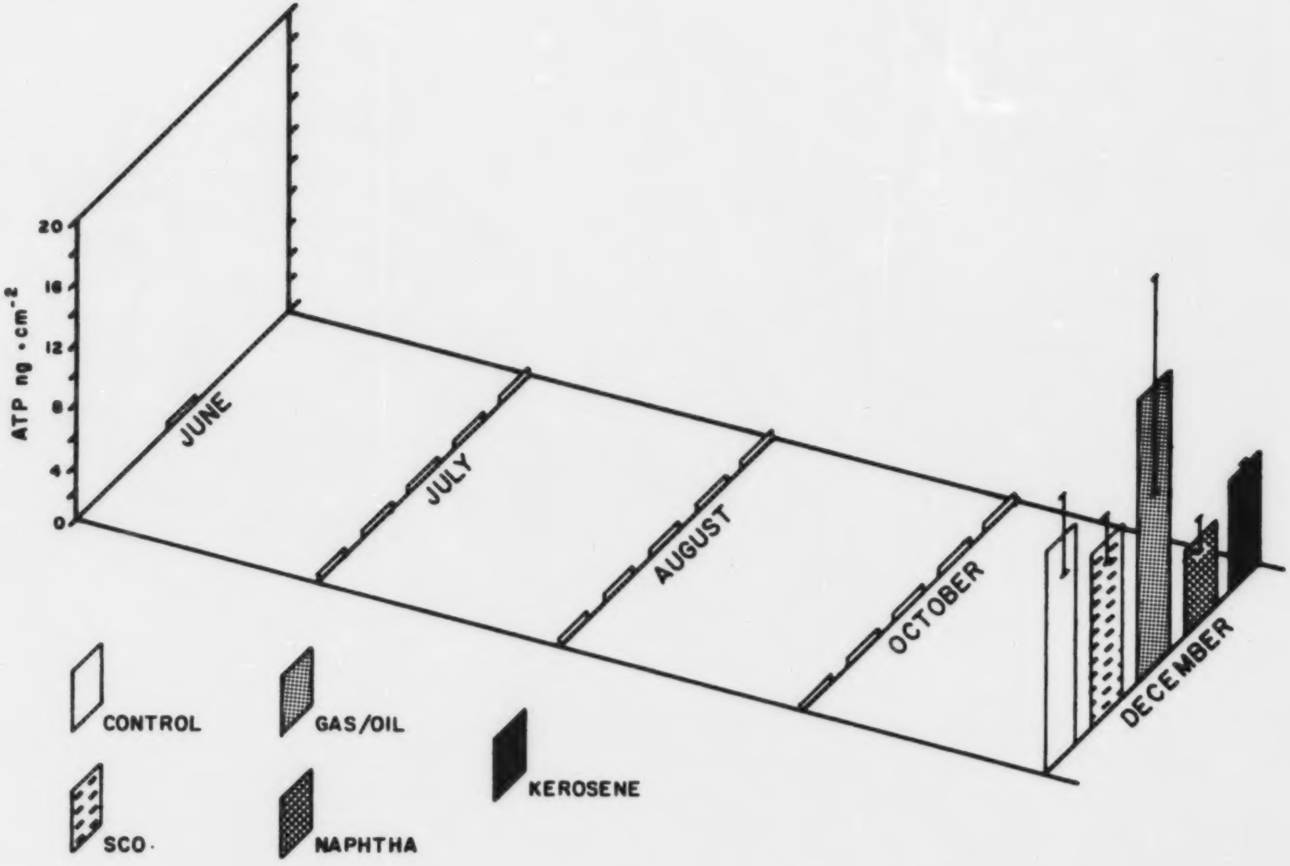

Figure 38. Mean ± standard error for ATP levels, based on five replicate samples (SCO--synthetic crude oil).

Table 33. Chlorophyll *a* in μg•cm^{-2} on control and oiled limestone bricks over time, mean ± standard error.

Date	Control	Synthetic Crude Oil	Gas-oil	Naphtha	Kerosene
28 June (pre-treatment)	0.75±0.19 (n=5)	N/A[a]	N/A	N/A	N/A
26 July	3.15±1.2 (n=4)	3.46±1.12 (n=4)	11.58 (n=1)	5.63 (n=1)	10.88 (n=1)
23 August	0.46±0.07 (n=6)	0.42±0.07 (n=6)	0.24±0.04 (n=3)	0.32±0.06 (n=3)	0.22±0.05 (n=3)
26 October	19.03±0.99 (n=6)	16.34±2.58 (n=6)	10.94±3.70 (n=3)	14.77±5.07 (n=3)	15.82±0.29 (n=3)
6 December	12.73±0.43 (n=6)	15.66±2.30 (n=6)	14.81±3.21 (n=3)	15.15±3.05 (n=3)	13.14±1.22 (n=3)

[a] N/A = Not applicable.

Table 34. Comparison of the means of the levels of chlorophyll between the control and synthetic crude oil and its three components[a].

Samples	July		August		October		December	
	t	df	t	df	t	df	t	df
Control vs. Synthetic Crude oil	0.192^{NS}	6	0.467^{NS}	10	0.318^{NS}	10	1.233^{NS}	10
Control vs. Gas-oil	N/A		2.157^{NS}	7	1.687^{NS}	7	0.798^{NS}	7
Control vs. Naphtha	N/A		1.366^{NS}	7	0.585^{NS}	7	0.799^{NS}	7
Control vs. Kerosene	N/A		2.360^{NS}	7	0.526^{NS}	7	0.423^{NS}	7

[a] N/A = Not applicable

NS = Not significant

ATP levels were below the detection limit of the technique employed (<1.0 ng·cm^{-2}) until December, when only naphtha and kerosene were significantly different from the controls (Figure 38, Table 35).

4.2.4 Discussion

Throughout the time of the experiments, although significant numerical differences emerged, there were no pronounced shifts in community structure on the oiled bricks as compared with the control bricks. The most striking occurred in July when there was a 3.74-fold increase in bacteria on synthetic crude oil-treated bricks, and the 2.0-fold increase on the bricks exposed to the oil components. As this was in accord with a similar increase found in Section 4.1.1, it seems reasonable to conclude that this is a response, at least in part, due to a stimulation of bacteria which degrade hydrocarbons. No increase in the amount of chlorophyll *a* was detected on the synthetic crude oil-treated bricks, but increases were noted on those treated with gas-oil and kerosene. However, such a response was not detected in the direct algal counts. From August onward, no significant difference in algal biomass as indicated by chlorophyll was detected; however, differences did appear with analyses of direct count data. The most marked differences between the control and oiled bricks were among the chlorophycean and rhodophycean algae, but the responses were extremely varied with time and the type of oil. Such a result is not surprising since several studies have demonstrated that the responses of algae are highly variable and dependent upon species and even strain-specific activity (Kauss et al. 1973; Pulich et al. 1974; Kauss and Hutchinson 1975; Parsons et al. 1976). However, it must also be pointed out that members of the Chlorophyta and Rhodophyta together composed only about 1% of the algal populations. The greater numerical proportion of algae were members of the Bacillariophyta and Cyanophyta, particularly the latter, and with these two groups, there were much smaller divergences from the control, generally within ±50%. Having said that, it is important to stress that the

Table 35. ATP in $ng \cdot cm^{-2}$ on control and oiled limestone bricks over time, mean ± standard error.

Date	Control	Synthetic Crude Oil	Gas-oil	Naphtha	Kerosene
28 June (pre-treatment)	< 1.0	N/A[a]	N/A	N/A	N/A
26 July	< 1.0	< 1.0	< 1.0	< 1.0	< 1.0
23 August	< 1.0	< 1.0	< 1.0	< 1.0	< 1.0
26 October	< 1.0	< 1.0	< 1.0	< 1.0	< 1.0
6 December	14.7±2.6	11.6±1.6	18.7±7.2	5.8±1.1	7.8±0.4

[a] N/A = Not applicable.

significance of any single component is largely unknown, be it a major or minor representative of that system. Again, no consistent responses were observed. A lack of response to oil additions was also demonstrated by Hanna et al. (1975) who found the addition of Norman Wells crude oil to a mixed community had little effect either numerically or in terms of ^{14}C productivity.

Since it has already been demonstrated, under different conditions (Section 4.1), that synthetic crude oil has a strong stimulatory effect, the question is posed, why did the weak response occur here only during the first month after the exposure? It may be due, in part, to the mode of presentation of the oils to wet, colonized substrates which may have allowed for only a small amount of the oil to become incorporated into the biological film. This aspect requires further investigation as spills are less likely to be of an instantaneous nature as was modelled in this study, but are more likely to extend to several hours or days of exposure.

The few significant responses by macrobenthos to the oils indicate that there were no great shifts in community structure, either positive or negative, in response to the oil contamination. It is doubtful that the effects, even where there were numerically significant responses, represented substantive shifts in the macrobenthic responses. *Simulium* showed a significant avoidance of the lighter oils (naphtha and kerosene) in July, which may be a reflection of difficulties in attachment to the oily surfaces. Other studies with light, refined hydrocarbons have demonstrated toxic effects to benthos. Bugbee and Walter (1973) demonstrated that aviation gasoline spilled into a river caused losses of Ephemeroptera and Plecoptera while *Orthocladius* was classified as a "resistant" organism to the oily residue. In another study, Rosenberg and Wiens (1976) classified *Eukiefferiella* as a strong to moderate positive responder to oils, a tendency which was only evident on our kerosene-contaminated bricks in August.

It is possible that this experimental study allowed for rapid losses of the oil from the substrate, either by microbial action and/or by erosion. As such, the effect of the oil, certainly as measured by macrobenthic responses, was greatly reduced after August. When considered in perspective with Section 4.1, it is apparent that microbial responses occur rapidly on oiled substrates, activity which is followed, apparently, by the macrobenthos. However, after about one month, great changes in macrobenthic communities were not evident.

4.3 THE ABILITY OF EPILITHIC AND SUSPENDED MICRO-ORGANISMS OF BROWN-WATER RIVERS TO DEGRADE SYNTHETIC CRUDE OIL

4.3.1 Introduction

A considerable number of studies have been concerned with the biodegradation of hydrocarbons by micro-organisms in terrestrial soils (Jobson et al. 1972; Westlake et al. 1974) and in marine and estuarine waters and sediments (Walker et al. 1975, 1976; Atlas 1975). Studies on freshwater systems, however, are extremely limited (Horowitz and Atlas 1977). In this study, the objective was to examine the potential capacity of the suspended and epilithic communities of two brown-water rivers, the Muskeg and Steepbank, to biodegrade synthetic crude oil which is the end product of the oil sands extraction-upgrading process. These rivers are especially interesting as one of them, the Steepbank River, cuts down into oil sands deposits toward its confluence with the Athabasca River. Samples were taken above and below a section of the Steepbank River, where there was considerable contamination of the substrate with oil sands, to determine if microbial communities, which have been in intimate contact with naturally occurring hydrocarbons, have an enhanced ability to degrade oil.

As a reference standard of microbial degradation of synthetic crude oil, a culture was included which had been through several transfers on a medium which contained synthetic crude oil as the sole source of carbon.

4.3.2 Materials and Methods

Water and sediment samples were collected from three sites in the Muskeg River, 8 km from the confluence with the Athabasca River, at the confluence of the North Steepbank River and of the Steepbank River (upper Steepbank), and 0.5 km from its confluence with the Athabasca River (lower Steepbank). The latter site was in the part of the river where it cuts a channel through oil sand deposits and thus had a long history of exposure to hydrocarbon material, whereas the other two sites had normal, rubble substrates. A more detailed description of the sites can be found in Section 4, where they are designated 1A, 6, and 7, respectively (Figure 2).

On 9 December 1977, at each station, 250 mL of water were collected in a sterile bottle and rocks from an area of approximately 0.04 m^2 were collected and flown back to the Mildred Lake field laboratory. There, the rocks were scrubbed into 500 mL of filter sterilized water and the resulting suspension transferred to a sterile bottle. The epilithon suspension and water samples were held at $2^{\circ}C$ until shipped to the Northern Forest Research Centre, in Edmonton, where the experiments were set up. Three days elapsed between the collections and the beginning of the laboratory experiments.

The "water" samples were set up by dispensing 150 mL of either Steepbank or Muskeg river water (which had been previously frozen to $-50^{\circ}C$) into 500 mL Erlenmeyer flasks, which were then autoclaved for 20 min. After cooling, 50 mL of the respective water sample (not previously frozen) were added to each flask. The "sediment" samples were set up similarly, except that 190 mL of previously frozen water were added, to which 10 mL of corresponding sediment samples were added. The total volume added to all flasks was 200 mL.

The negative controls (two for the Steepbank River at each temperature, and one for the Muskeg River at each temperature) were set up by adding 200 mL of a previously frozen water sample to each flask and autoclaving it. Positive controls (the same number of flasks as the negative controls) had 190 mL of previously frozen water sample added per flask and were autoclaved. After cooling, 10 mL of a culture known to use synthetic crude oil were added to these positive controls. This mixed culture was maintained in constant exposure to synthetic crude oil in the laboratory. Another set of positive controls (two for each river) was also set up, in the same manner as above, except that filter sterilized, rather than autoclaved, river water was used. A comparison of the results obtained for the two sets of positive controls would show if there is any difference in biodegradation capabilities between filter sterilized and autoclaved samples.

Synthetic crude oil was added in the ratio of 0.2 mL per 200 mL of culture. All flasks were also supplemented with 2 mL of a nitrogen/phosphorus solution containing 7% K_2HPO_4, 3% KH_2PO_4, and 10% NH_4NO_3 (all w/v).

All samples were incubated at $4^{\circ}C$ or $20^{\circ}C$ on gyrotary shakers having a 1 in (2.54 cm) eccentricity at 250 RPM. The length of the incubation period varied and the specific times used are shown in Tables 36 and 37. At the end of the incubation period, the flasks were removed from the shakers and acidified with 50% HCl (ca. 1 mL) to a pH <1, which was checked with pH paper. The saturates of synthetic crude oil were then extracted from the flasks via a floatation method of Dr. A. Jobson (Alberta Research Council, Edmonton; unpublished method). Five millilitres of pentane were added to the flask, the flask was tightly stoppered, inverted several times to wash any hydrocarbon from the walls of the flask, and then shaken on a rotary shaker at 250 RPM for 25 min. The stopper in the flask was then exchanged for a bung with two glass tubes, the pentane extract was floated to the top of the flask by adding water to the flask through one of the tubes. A 10 µL GC syringe was then inserted into the second glass tube and 2 µL of the pentane extract were removed, injected into a Varian gas-liquid

Table 36. Biodegradation of synthetic crude oil by samples incubated at 4°C.[a]

Sample	5 weeks incubation	7 weeks incubation
MUSKEG RIVER		
Autoclaved water control	-	ND
Synthetic crude oil utilizer	+++	ND
Filtered synthetic crude oil utilizer	ND	+++
Midstream sediment	++	+++/+++
Water	-	+++/+
STEEPBANK RIVER		
Autoclaved water control	-	-
Synthetic crude oil utilizer	+++	+++
Filtered synthetic crude oil utilizer	ND	+++
Lower sediment	+++	+++/+++
Lower water	+	+++/++
Upper sediment	+++	+++/+++
Upper water	-	+++/+

[a] ND = not done

- = no biodegradation

+ = biodegradation starting as evidenced by slight reduction of low molecular weight n-alkanes

++ = significant biodegration; reduction of most or all saturate peeks

+++ = "complete" biodegration; little or no peaks remain

Table 37. Biodegradation of synthetic crude oil by samples incubated at 20°C.[a]

Sample	4 weeks incubation	5 weeks incubation
MUSKEG RIVER		
Autoclaved water control	-	ND
Synthetic crude oil utilizer	+++	ND
Filtered synthetic crude oil utilizer	ND	++
Midstream sediment	+++	+++/+++
Water	+++	+++/+++
STEEPBANK RIVER		
Autoclaved water control	-	-
Synthetic crude oil utilizer	+++	+++
Filtered synthetic crude oil utilizer	ND	+++
Lower sediment	+++	+++/+++
Lower water	+++	+++/+++
Upper sediment	+++	+++/+++
Upper water	+++	+++/+++

[a] ND = not done

- = no biodegradation

+ = biodegradation starting as evidenced by slight reduction of low molecular weight no-alkanes

++ = significant biodegradation; i.e., reduction of most or all saturate peaks

+++ = "complete" biodegradation; little or no peaks remain.

chromatograph, and the following temperature program was initiated: 50°C for 2 min from the time of injection, then a linear temperature program from 50 to 300°C at $10^{\circ}\cdot\text{min}^{-1}$.

The GLC used for the analysis was a Varian aerograph fitted with two separate columns and flame ionization detectors, allowing two samples to be analyzed simultaneously. The columns were 20 ft x 1/8 in O.D. (0.020 in wall) stainless steel packed with 3% SE-30 on Chromosorb W, with nitrogen as the carrier gas at $15\ \text{mL}\cdot\text{min}^{-1}$. The detector gasses were hydrogen and compressed air flowing at 30 and $300\ \text{mL}\cdot\text{min}^{-1}$, respectively. The output was recorded on Hewlett-Packard recorders, using a chart speed of $1.25\ \text{cm}\cdot\text{min}^{-1}$.

4.3.3 Results

It is important to emphasize that this technique primarily monitors degradation of the saturate fraction only and is essentially qualitative. Four degrees of degradation are reasonably distinct and examples are given in Figure 27. The negative control (-) represents the saturates remaining after physicochemical processes, presumably mainly evaporation. A + indicates only a slight change from the control. To see this difference, it is necessary to compare the relative peak heights of two couplets of peaks occurring in the early part of the GLC trace (Figure 39). These couplets are located at retention time 14.5 and 16.5 min. The first peak is an n-alkane and shows a reduction in height in relation to the small peak to the right (probably on isoprenoid). The +++ represents "complete" biodegradation of the saturate fraction with a substantial reduction of all the peaks occurring. The ++ represents an intermediate stage between + and +++.

Results of the study are presented in Tables 36 and 37. The positive controls all show extensive biodegradation. Only one sample (Table 37, which was a filtered synthetic crude oil utilizer incubated at 20°C for 5 wk) showed a negligible difference between the GLC traces of the filtered and autoclaved positive controls.

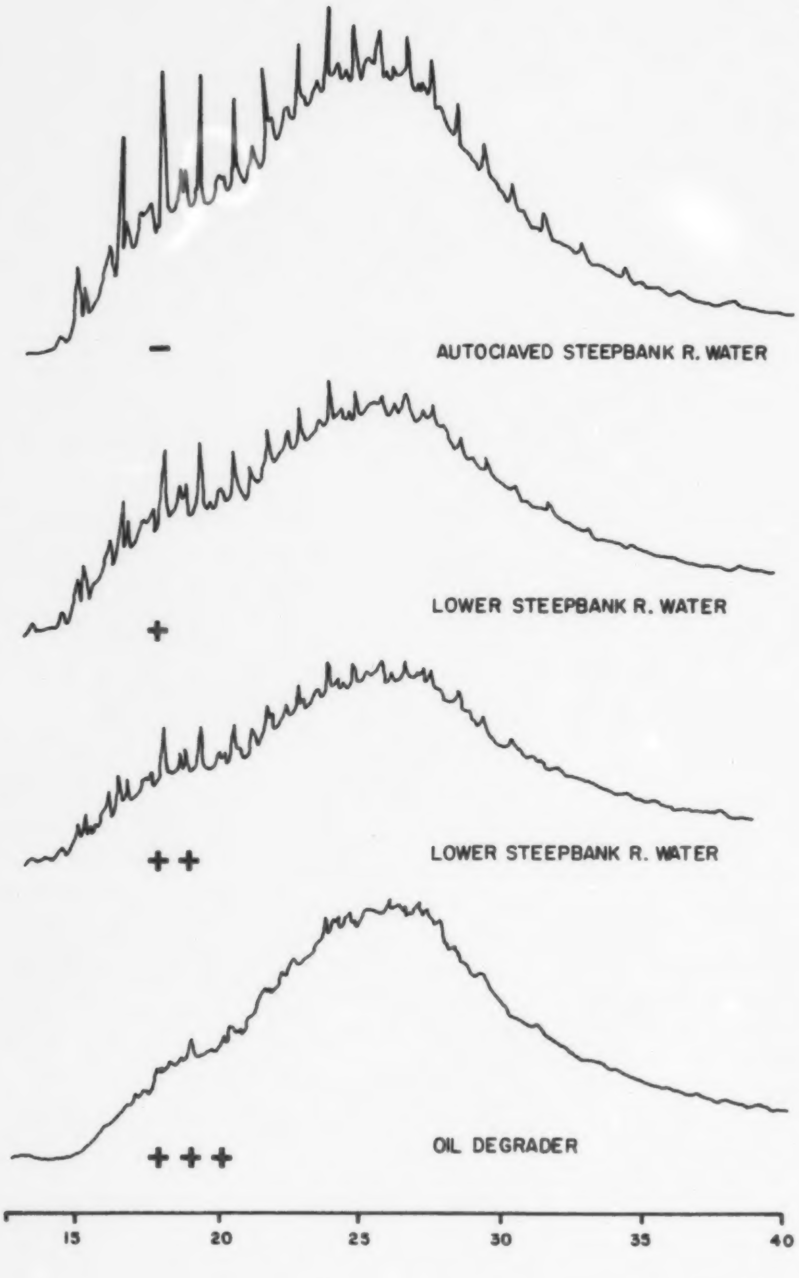

Figure 39. The degrees of oil degradation evident after incubation. The standard demarcations used in (-, +, ++, +++) are as indicated in Tables 36 and 37.

This confirmed that the autoclaving process was apparently not altering the chemical composition of the water which might have affected the bacterial activity.

The results of the experiment at 20°C show that epilithon and water from both the Muskeg and Steepbank rivers completely degrade the saturate fraction of synthetic crude after 4 wk incubation. There were no detectable differences between samples for the two rivers, epilithon and water, or the two different incubation times.

The results of the experiment at 4°C are a little more complex. These show a marked difference between the water and epilithon samples, particularly after only 5 wk incubation. At this time, the water samples show no biodegradation (or very little for lower Steepbank River water) in comparison with complete or nearly complete biodegradation for the sediment samples of both rivers. The water samples are clearly capable of biodegradation of synthetic crude oil since, after 7 wk, they show considerably more change than after only 5 wk. Given enough time, it is likely that all samples would show "complete" biodegradation although the rate at 4°C is significantly slower than at 20°C.

4.3.4 Discussion

These data quite clearly demonstrate the presence of oil-degrading micro-organisms in both the upper and lower reaches of the Steepbank River and in the Muskeg River, and that degradation takes place more quickly at 20°C than at 4°C. Also, on the basis of the experimental series at 4°C, there is a suggestion that the epilithic material may have a greater capacity for oil degradation than the suspended material. This may simply be a reflection of the size of the initial innoculum of bacteria. In the case of the epilithic material, this was around 3×10^9 bacteria, and for the suspended material, from 0.6 to 3×10^7. However, an approximate 100-fold difference is also found when comparing the numbers of epilithic bacteria$\cdot$cm^{-2} with the number$\cdot$mL^{-1} occurring naturally in these rivers (Lock et al. in prep.), thus, the initial starting ratios of bacteria may not be too different.

As noted earlier, biodegradation here refers only to the saturates. Atlas (1975) was able to demonstrate that the paraffinic aromatic and asphaltene fractions and even some branched compounds were also subject to degradation. However, he found that, even in those oils most susceptible to degradation, a residue of 20% seemed to be resistant. Walker et al. (1976), working with estuarine sediments, found that the total degradation South Louisiana crude oil, by cultures having reached a stationary phase, was 18% for innocula from a clean area and 47% for those from an oil-contaminated area. They also observed considerable degradation (30.7%) of the aromatic fractions by the sediment from the oil-contaminated area. It may be that, on following the degradation of a wider spectrum of the hydrocarbons composing synthetic crude oil, the micro-organisms associated with oil sand in the lower reaches of the Steepbank River would be seen to have a greater ability to degrade the more resistant components than the cleaner Muskeg River. Indeed, it would be valuable to know if the rivers of the AOSERP study area are unique in their considerable ability to degrade the saturate fraction and possibly other more resistant organic components of oils.

4.3.5 Discussion

Moore and Dwyer (1974) identified five basic classes of effects of oil on marine organisms, many of which apply to freshwater organisms: (1) direct lethal toxicity; (2) sub-lethal disruptions; (3) effects of direct coating (on animals); (4) incorporation of hydrocarbons; and (5) habitat alterations, especially the character of the substrate. Of these, only the latter two seem directly applicable to the present longer term studies. Moore and Dwyer (1974) noted that there was considerable evidence for marine toxic responses to oils caused primarily by the lower boiling aromatics (the low boiling point usually corresponds with increasing solubility), of which the more soluble aromatic fractions are consistently implicated as the toxic agents. Refined hydrocarbons, such as No. 2 fuel oils, are, therefore, predicted to cause greater mortalities than equivalent amounts of crude oils, because of the greater relative amounts of medium boiling aromatics such as naphthalene. Such considerations are, however, of limited use in longer term, substrate-benthos interactions since degradation or erosion of toxic fractions may occur relatively quickly, especially in running waters. Also, the apparently disparate effects of oils in waters are partly explained by the differing chemical nature of various hydrocarbons and their changing composition over time. For instance, Busdosh and Atlas (1977) indicated that Arctic amphipods were very susceptible to paraffinic oil components of crude oil and diesel fuel, but that the asphaltic fraction of the crude oil, which remained after degradation or weathering, was not lethal to the amphipods. Indeed, the amphipods appeared to browse on the residues, an occurrence which has also been reported for other oceanic crustaceans (Thomas et al. 1977). That hydrocarbons may rapidly disappear from the surface of standing waters (by solution and principally volatilization) has been demonstrated by Miller et al. (in press) and Snow and Rosenberg (1975a, 1975b). Losses of 10% and 30 to 40%, respectively, were demonstrated in the first day after experimental spills. Lysyj and Russel (1974) found that, after a period of stabilization, ranging from 40 to 24 d, an acceleration of the dispersion of oil into water may occur which is associated with chemical changes in the composition of the film of oil, particularly the water-insoluble petroleum fraction. They found substantial quantities of aromatic compounds, including phenolics, in subsequent fuel extracts. In the oiled bricks of the present study, the toxic phenolics or aromatics in the oils may have leached out into the moving column of water. Much of the literature on the effects of hydrocarbons in running waters is difficult to interpret, due to a lack of characterization of the oils or of the benthic communities, especially microbial and macrobenthic interactions. For instance, Ludzak et al. (1957) found that benthic responses to oil refinery and petrochemical effluents were correlated with deposits of "oily sludge" and Hoehn et al. (1974) found that, after an experimental spill of No. 2 fuel oil, benthic invertebrates did not find the substrate suitable for colonization. Similarly, Bengtsson and Berggren (1972) found oily sediments from a stratified Swedish lake toxic to *Chironomus* larvae in laboratory tests, but field sampling indicated the presence of Tubificidae (*Limnodrilus*) in the sediments, whereas, McCauley (1966) found that *Gammarus*, *Agrion*, and *Dugesia* were not present in areas of a stream contaminated with bunker oils, while *Tubifex*, *Chironomus* larvae, Nemata, and Hirudinea remained. Tubb and Dorris (1965) found that benthic populations of *Chironomus*, *Harnischia*, and *Tanypus* in refinery effluent holding ponds increased in successive ponds which indicated that certain critical concentrations of the oily effluents may have exerted toxic effects. While such descriptive studies are useful in assessing specific occurrences of oil pollution, they do not address the more substantive questions of microbial-macrobenthic interactions, or of the dynamics of recovery of such communities from hydrocarbon inputs.

Certainly, it has been known for some time that microbial communities may flourish either as a direct response to stimulation by hydrocarbons, or as a result of decreased grazing pressure from benthic herbivores which may have been eliminated by hydrocarbon toxicity. In 1965, Hynes demonstrated that, while the sewage fungus *Sphaerotilus* flourished in a Welsh river downstream from an organic effluent (mostly phenolic), many invertebrates (Ecdyonuridae, Plecoptera, and Elmidae) were absent. Abelson (1977) noted that a wide variety of micro-organisms metabolize or detoxify hydrocarbons, the straight-chain molecules being most susceptible to attack, and Shindler et al. (1975) and Atlas (1977) found increases in total bacterial numbers in Arctic freshwaters treated with oils. Similarly, Snow and Rosenberg (1975a, 1975b) also found that the biomass of phytoplankton and periphyton increased in oiled Arctic lakes, particularly the blue-green algae which became dominant.

It is clear from the laboratory studies, that the micro-organisms of the Steepbank and Muskeg rivers, and probably other rivers in the Athabasca Oil Sands area, are able to degrade at least the saturate fraction of synthetic crude oil. Although the degradation occurred more quickly at higher temperatures, the rates are as yet undefined and further work of a quantitative nature is needed to resolve this question.

The responses of the Muskeg River benthic communities to the additions of oil will now be considered. As such, the findings of the first experiment (Section 4.1) are quite different from the second (Section 4.2). The former demonstrated the effects of an oil spill to a river bed during a period of low water, followed by increased water levels. Here, the effects of the oil were detectable for, at least, 4 wk. The oil which was in intimate contact with the limestone surface led to populations of bacteria greater than on the controls and, in the light, to algal growth which in turn supported significantly more macroinvertebrates, particularly the Chironomidae and Naididae (Table 24). It is not possible to say to which populations (the bacteria or the algae) of the macrobenthos were responding in the light, since both were enhanced on the oiled substrata in the light (Figure 31). However, since specific responses by macrobenthos for microbenthic species are known (Barlocher and Kendrick 1974), it is conceivable that there is an enhancement of an algal or microbial population which is specifically responding to the oil, which is, in turn, specifically attractive to certain species of macroinvertebrates.

In the second experiment, the bricks which were allowed to colonize before exposure to the oils (Section 4.2) simulated a spill into running waters. Here, established benthic communities, including micro-organisms which were seen to be capable of oil degradation in the laboratory, could prevent contact of the oil with the substrate and could immediately begin assimilation of the oil.

In each of these experiments, the macroinvertebrates were probably able to colonize the contaminated bricks rather more quickly than would have been the case for a true oil spill to a river. In the latter case, there probably would not be a large, unaffected, reservoir of healthy animals potentially able to recolonize the oiled substrata, as in this experiment.

Little differences was found between the controls and the oiled bricks after one month, save for the total count of bacteria, which was considerably higher on the oil-treated bricks (2 to 3.7-fold) (Section 4.2). After July, differences between the control and oiled bricks were not apparent, with the exception of the number of Rhodophycean and Chlorophycean algae. However, these represented a maximum of only 2.5% of the total algal population and were generally less than 0.5%. This was also generally true for the macrobenthos as well. While it is true that some statistically significant numerical differences were noted in July and August, there were no pronounced or consistent shifts in the general composition or distributions of the fauna as compared with the control samples. Further, the disappearance of significant numerical differences by October indicates that any effect of the oil was negligible after August.

Although *Eukieffriella* showed a significantly positive response to kerosene-contaminated bricks in August, it is interesting that other known responders to oils, such as the Orthocladiinae and *Cricotopus bicinctus* showed no pronounced response during the times (July and August) when the oil residues should have been maximal. This indicates that the community responses, toward or away from the oil contamination, were rather minimal, or at least short lived. As there were many significant responses (within 30 days) to the first oil experiment (Section 4.1), in which dry bricks were soaked in synthetic crude oil and then placed in the river, it seems reasonable to conclude that the microbial or algal responses to these light oils are rapid, largely occurring within 5 wk of the initial contamination.

It is appropriate to consider the possible mechanisms for the increases of bacteria, algae, and some macroinvertebrates associated with the oil during the first weeks of the exposure. Suggestions and evidence for increased algal standing crop in the presence of oil, being due to a reduction of grazing pressure brought about by the elimination of zoobenthic grazers by toxic fractions of the oil, has been presented by several authors (Rosenberg and Wiens 1976; O'Brien and Dixon 1976; Federle et al. 1977; Miller et al. in press). However, Rosenberg and Wiens (1976) suggested that this was unlikely in their study, as herbivorous Chironomidae were found to be unaffected by crude oil additions and suggested that nutrients from the oils stimulated algal growth which attracted Chironomids, particularly Orthocladiinae. Rosenberg et al. (1977) further suggested that *C. bicinctus* and *C. mackenziensis* responded positively to oil-contaminated substrates because of effects following from algal blooms, such as amelioration of toxic effects or the provision of food. In the present study, there was a greater abundance of grazing species on the oiled bricks in sunlight than on the control (Section 4.1), which would negate the contention that a release of grazing pressure resulted in the increased microbial biomass as compared with the controls.

However, such a phenomenon has been observed in standing, fresh waters (Miller et al. in press) and it would, therefore, appear that community responses to oil contamination may be quite different in standing and running waters. It would appear that in running waters, a microbial response to the oil is the prime determinant which sets off community change. The experiments suggest that refined, light oils, either directly or indirectly, stimulate the growth of microflora. There are several possible mechanisms which could account for this growth, which is apparently followed by macrobenthic organisms:

1. The oil could act as a carbon source to heterotrophic organisms which, in turn, act to accelerate regeneration of nutrients in the epilithon;
2. The oil could stimulate nitrogen-fixing bacteria (Shindler et al. 1975) which, in turn, cause an enhanced supply of nitrogen to the epilithon;
3. There could be growth-stimulating compounds present in the oils, such as trace elements or compounds which regulate growth (Baker 1971); and
4. The oily surface could physically enhance the trapping of nutrients by sorptive processes (Marshall 1976).

Only point 3 does not assume that nutrients would be limiting bacteria and algal growth, an assumption which needs testing.

The general contention that microbial responses to oil contamination, possibly caused by any of the above-noted points, govern the responses in the macrobenthic communities is favoured. As such, it is felt that the traditional concept of indicator species needs greatly to be expanded to include detailed considerations of the prime determinants, such as the lower trophic levels for subsequent macrobenthic responses. With the relatively light, refined, crude oil used in the present experiments, it appears that the microbial response, and the subsequent macrobenthic response, to the oils is very rapid, largely occurring within a month of the [illegible]mination. This is further supported by the laboratory findings of rapid, active microbial degradation of synthetic crude oil by microbes from the Muskeg, and especially Steepbank, rivers. It is important to note that the present findings are based on biomass data and it is desirable to conduct further investigations using productivity as a measure. While there are no data on the erosion or disappearance of the oils from the substrates, presentation of oils to dry, uncolonized, substrates may allow for deeper absorption of the oil into such limestone and may provide a more stable environment for continued microbial growth down into the oil, limestone, substrate.

Obviously, massive oil spills to running freshwaters will have an initial detrimental effects on fishes and benthos (Bugbee and Walter 1973). However, the subsequent degradation or erosion of the oil by natural physical or biotic processes may allow for a more rapid recovery than was previously thought, at least for light oils. Here, the objective has been to simulate acute short-term spills of oil to a river and the concern was not about the initial toxic response but about the time of recovery and nature of the response by benthic communities. As such, the studies could be categorized as simulating a medium to severe spillage of oil to a river by the classification scheme proposed by the European Petroleum Organization (1974). Rosenberg and Wiens (1976) point out the need for quantitative data on the amount of oil spilled onto the aquatic substrates, but they offer no clear advice as to how to make comparative measures for such contamination. Our approach was to carefully standardize the technique to permit replication in future for more detailed studies, particularly those with low-level chronic contamination of oil in rivers.

4.4 A PRELIMINARY STUDY OF STREAM RECLAMATION TECHNIQUES FOR OIL SANDS DEVELOPMENTS

4.4.1 Introduction

One of the principal disturbances to streams in mining areas of the AOSERP study area will result from the diversion or surface mining of river valleys which overlie oil sands deposits.

In the case of stream diversions, it is presumably important to design such channels so that there is not only an efficient flow of water but also so that the biological productivity within the channels is optimized. Such diversion channels could represent important recreational or aquatic habitat areas during and following oil sands development. In the case of possible mining activities in river basins, such as the Beaver River area within the Syncrude Canada Ltd. lease, a principal objective after development will probably be to return the affected areas to their original levels of productivity. Therefore, stream reclamation techniques are of importance in the planning of subsequent, man-made drainage basins or portions thereof.

The study described herein examined several types of substrate, ranging in size from tailings sand up to large cobbles, which would probably be available for use in the construction of diversion channels or in plans for future reclamation techniques. It was hoped to obtain a preliminary comparison of the nature of macrobenthic communities which became established upon such substrates over a reasonable length of time. Such studies allow for a more precise planning of the structure and use of aquatic habitats in reclaimed channels or in diversion canals and allow for more detailed research.

4.4.2 Materials and Methods

Two sets of experimental channels, patterned after that used by Dejoux (1975), were constructed from 1.9 cm (3/4 in) plywood coated with clear urethane varnish. Each channel was 1.2 m long and 30 cm wide, and each set consisted of six parallel channels. Two channels of one set were each filled to a depth of 10 cm with sand from the Muskeg River (river sand), from a nearby, dry hillside (bank sand), or tailings sand obtained from Great Canadian Oil Sands Ltd. (GCOS). The distribution of particle sizes of each type of sand is given in Table 38. The lower 10 cm of each end of these channels were covered with 2.5 cm mesh wire screen which supported a 1 mm mesh nylon screen to hold the sand in place but allowed the passage of water.

Five cubical, open baskets (15 x 15 x 15 cm), constructed of 2.5 cm wire mesh with a bottom screen of 202 µm nylon mesh, were each placed 5 cm apart along the centre of four channels of the second set. Two channels of this set were each filled to a depth of 15 cm with river gravel shoveled from the streambed, bank gravel quarried from a nearby road cut (in both cases filling the baskets simultaneously), or two layers of rounded cobbles (8 to 10 cm in diameter) gathered from nearby hillsides. Both types of gravel consisted of angular particles of Devonian limestone up to 5 cm in maximum length as well as particles in the sand and clay size ranges (Cummins 1962). The cobbles were smoothly rounded and were found in glacial till. The lower 15 cm of each end of these channels was screened as in the sand set. After the channels were filled, the buoyancy of both sets was adjusted to neutral using sheets of styrofoam insulation fastened under the channels. Each set was moored to steel posts driven into the riverbed so that the surface of the substrates was about 15 cm beneath the water surface.

The gravel channels were put in place on 25 June 1977 near midstream just above Site M-1A (Figure 2) where the flow of the river was nearly laminar at speeds of about 30 $cm \cdot s^{-1}$. The sand channels were set up on 8 July in an adjacent pool where current speeds were less than 5 $cm \cdot s^{-1}$. Five samples were collected from each channel

Table 38. Particle size range distributions (%) on the types of sand used in the channels.

Particles less than: (mm)	River Sand	Bank Sand	Tailings Sand
2.000	100.0	100.0	100.0
0.841	95.1	99.9	99.4
0.420	27.6	97.3	97.7
0.177	7.4	26.6	47.6
0.149	5.8	13.7	32.5
0.125	4.9	7.7	20.3
0.074	3.6	2.7	6.8

on 30 September. Sand was sampled by pressing a metal cylinder (5.6 cm in diameter, 12 cm long) to the bottom of the channel and digging away the surrounding sand so that a flat plate could be slid under the cylinder and the sample lifted out. The gravel channels were sampled by lifting each basket quickly into a 202 µm mesh net held just downstream. Each cobble sample consisted of two stones from each of the top and bottom layers lifted into a net held just downstream. (The volume and surface area of the channel occupied by four cobbles approximated that occupied by each basket.) Each sample was emptied into a bucket of river water and organisms were removed from it by scrubbing the large stones with a vegetable brush, agitating, and decanting through a 180 µm sieve. The agitating and decanting procedure was repeated eight times and the concentrated organisms were immediately preserved in 10% formalin.

The animals were sorted from the samples under 10X magnification in the laboratory and placed in 70% alcohol. Associated algae and organic detritus from each sample were air-dried at room temperature for 2 wk and then weighed to the nearest milligram. All the animals from the samples of sand were identified and enumerated at the taxonomic levels listed in Appendix 6.1. Samples from only one river gravel, one bank gravel, and one cobble channel were analyzed. Animals in these samples were identified and enumerated as in the sand samples except that Oligochaeta and Chironomidae were identified from only two samples from each channel. After identification, the animals from each sample were dried at $40^{\circ}C$ for 2 h and weighed to the nearest 0.1 mg. It should be emphasized that this technique yields only relative, not absolute, estimates of biomass.

Since the variances of sample counts and weights tended to be large, all the values were transformed as $\ell n\ (n + 1)$ (Bartlett 1947) before testing for significance of differences between geometric means using Student's t test. To avoid the encumbrance of unnecessarily large numbers, sample counts and weights were used to compare standing stocks between substrates. These may be converted to values per square metre by dividing by 0.00985 (sand samples) or 0.0225 (gravel and cobble samples).

4.4.3 Results

At the end of the experiment, on 30 September, each of the sand channels contained a dense growth of filamentous and epiphytic algae which completely covered the sand and was attached to the walls of the channels. The upper surfaces of the cobbles and gravels in the channels were almost completely covered with *Cladophora glomerata* and *Ulothrix zonata* and considerable quantities of leafy and woody detritus were caught between the stones. The density of macroalgae seemed to be least in the channel filled with bank gravel.

Mean numbers of animals per sample in major taxonomic groups, total benthos, biomass, and the weight of algae and detritus found on each substrate are summarized in Table 39. The variety, number, and weight of invertebrates were greatest on river sand and river gravel as compared with the other substrates. Chironomid larvae numerically dominated the fauna on all substrates and accounted for 60 to 75% of the total invertebrate biomass on sand but less than 10% on the coarser substrates. On gravel and cobbles, larger insects, such as mayflies, stoneflies, caddisflies, and Sphaeriidae (on river gravel), accounted for most of the invertebrate biomass.

Samples from both bank sand and tailings sand contained significantly fewer taxa than did river sand (Table 40) but the composition of the fauna in each channel was essentially the same. The total number of individuals per sample and the density of chironomids were lower on tailings sand than on river sand, but no statistically significant differences were observed between bank and tailings sand even though only about half as many chironomids were found on tailings sand. The total standing stocks were: river sand, 23 300 animals$\cdot$m^{-2} (0.14 g$\cdot$m^{-2}); bank sand, 16 870$\cdot$m^{-2} (0.08 g$\cdot$m^{-2}); and tailings sand, 10 792$\cdot$m^{-2} (0.11 g$\cdot$m^{-2}).

The fauna of coarser sediments was much more abundant: river gravel, 157 000 animals$\cdot$m^{-2} (21.18 g$\cdot$m^{-2}); bank gravel, 33 600$\cdot$m^{-2} (6.03 g$\cdot$m^{-2}); and cobbles, 97 000$\cdot$m^{-2} (0.67 g$\cdot$m^{-2}). Both the number of individuals and biomass were significantly different between each pair of substrates (Table 40). About 50% of the invertebrate biomass found on river gravel consisted of Sphaeriidae, a group which was

Table 39. Mean numbers and weights of macrobenthos and organic matter on experimental substrates.

Taxon	Numbers			Numbers (mg)		
	RS	BS	RS	RG	BG	CO
Oligochaeta	8.8	7.0	6.1	117.1	53.5	11.7
Crustacea	37.3	22.8	25.6	30.1	30.8	45.9
Ephemeroptera	1.5	2.4	4.0	134.0	84.9	53.0
Plecoptera	0	0	0	106.0	21.7	13.0
Trichoptera	1.4	1.1	2.9	52.3	13.6	30.7
Chironomidae	181.7	114.2	58.9	2111.6	496.4	2049.2
Total, non-Chironomidae	56.8	41.8	34.4	1417.1	430.3	184.9
biomass (mg)	0.5	0.2	0.3	468.1	133.3	14.0
Total benthos	229.5	166.2	106.3	3532.6	757.0	2182.9
biomass (mg)	1.4	0.8	1.1	476.6	135.6	15.5
Number of Taxa	22.2	18.4	14.8	25.6+	18.6+	17.2+
Algae & Detritus (g)	0.918	0.480	0.234	2.880	3.301	0.993

[a]Symbols: + = exclusive of Oligochaeta and Chironomidae

RS = river sand
BS = bank sand
TS = tailings sand
RG = river gravel
BG = bank gravel
CO = cobbles

Table 40. Values of Student's t test from comparisons of pairs of substrates using ℓn-transformed counts and weights.

Taxa	Comparison BS/RS	 TS/RS	 BS/TS	 BG/RG	 CO/RG	 MG/CO
Oligochaeta	1.42	1.40	0.43	1.72	4.59[b]	3.07[a]
Crustacea	1.79	1.22	0.39	0.05	0.77	0.65
Ephemeroptera	0.04	0.77	0.69	1.49	2.80[a]	1.90
Plecoptera	0	0	0	3.50[b]	4.29[b]	0.84
Trichoptera	1.07	0.47	1.76	2.67[a]	1.83	1.74
Chironomidae	1.65	3.71[b]	1.89	3.65[b]	0.17	3.92[b]
Total, non-Chironomidae	1.35	1.71	0.69	4.91[b]	9.60[b]	3.66[b]
biomass	1.90	0.93	0.62	3.00[a]	5.36[b]	2.91[a]
Total benthos	1.65	4.18[b]	1.87	4.51[b]	2.87[a]	3.50[b]
biomass	1.63	0.68	1.00	3.01[a]	15.75[b]	4.81[b]
Number of Taxa	2.43[a]	3.74[b]	1.94	4.27[b]	3.53[b]	0.59
df	17	17	18	8	8	8

[a] $p < 0.05$

[b] $p < 0.01$

[c] Symbols:

BS = bank sand
TS = tailings sand
RS = river sand
BG = bank gravel
CO = cobbles
RG = river gravel

rarely collected on the other substrates. The much higher density of Chironomidae on cobbles as compared with bank gravel probably reflected the more luxuriant growth of filamentous algae on the former. While the numerical abundance of all Ephemeroptera, Plecoptera, and Trichoptera did not differ significantly between these two substrates, most of the individuals inhabiting the cobble channel were small species such as *Baetis* sp., *Oxyethira* sp., and very young *Oenopteryx fosketti* (Table 41). Relatively larger organisms (at least at the time of sampling), such as *Leptophlebia, Stenonema vicarium, Heptagenia* spp., *Hastaperla brevis,* and *Pteronarcys dorsata*, were more abundant in the gravel.

4.4.4 Discussion

Various materials are readily available for the construction of diversion channels prior to oil sands mining or the re-establishment of surface drainage in mined areas. Such materials include overburden, which is often sand, tailings sand, limestone rubble and gravel, and glacial till. The experiment just described was designed to assess the relative productivity, as estimated by standing stock and composition of the macrobenthic fauna, of these materials when used as stream substrates. Gravel and sand from the Muskeg River were used to simulate the natural communities found in unaltered streams.

Sand is, at best, a poor substrate for most benthic macroinvertebrates (Hynes 1970; Nuttall 1972; Luedtke and Brusven 1976) and the fauna which normally inhabit it in streams and rivers consists mostly of specialized chironomid larvae (Shadin 1956). Since both species accounted for only a small fraction (less than 5%) of the animals found in the sand channels, it seems reasonable to assume that most of the organisms were actually living among the filamentous algae which was growing on the side walls of the channels and lying on the sand. Similar algal mats were observed on sand, mud and gravel in backwaters of the Muskeg River in late summer, but these were transitory, being readily washed away or desiccated as a result of fluctuations in the discharge of the river. Thus, such algal mats may be

Table 41. Mean numbers of selected insects and results of t-comparisons between substrates.

	Mean Number			t		
	RG	BG	CO	BG/RG	CO/RG	BG/CO
Ephemeroptera						
Baetis	25.5	13.5	34.3	0.99	0.54	1.96
Leptophlebia	66.2	27.5	2.0	1.78	9.22[b]	6.54[b]
Heptagenia	18.5	11.7	3.5	2.24	3.18[a]	2.16
Stenonema	21.4	16.1	2.6	1.53	8.22[b]	6.37
Plecoptera						
Oenopteryx	28.3	4.1	8.8	2.83[a]	2.26	0.20
Pteronarcys	0.6	0.8	0.2	0.06	1.04	1.21
Hastaperla	68.5	12.9	2.0	3.81[b]	11.86[b]	4.28[b]
Trichoptera						
Hydroptilidae	7.6	2.1	25.9	2.29	2.50[a]	6.70[b]
Ceraclea	36.8	7.0	3.5	2.95[a]	7.03[b]	1.32

[a] $p < 0.05$

[b] $p < 1.01$

[c] Symbols:
RG = river gravel
BG = bank gravel
CO = cobbles

very important to the functioning of the stream ecosystem but do not necessarily reflect the relative productivity of the different types of sand used in this experiment. However, since little or no filamentous algae appeared to be present in the river sand at the beginning of the experiment and the development of such algal mats elsewhere in the river seemed dependent on a lack of current rather than the presence of a certain substrate, the smaller amount of algae on tailings sand suggests that tailings sand may inhibit growth of some algae. A possible reason for this is that tailings sand is known to contain few nutrients and some residual hydrocarbons (M. Korchinsky, Inland Waters Directorate, Calgary). These factors, in combination with the lower algal density, may also have contributed to the lower density of invertebrates as compared with river and bank sand.

While the masking effects of the algae preclude firm conclusions about the relative attractiveness of the three types of sand to invertebrates, it should be pointed out that two parameters, particle size and the amount of particulate detritus entrained by the sand, can be expected to be of greatest importance. The two are linked since coarser sands have larger interstitial spaces between the grains and are more readily shifted by currents of lower velocity (Schmitz 1961) which in turn leads to the incorporation of more detritus into the sandy riverbed. Percolation of water, dissolved gases, and nutrients are greater through coarser sand and the larger interstices provide more room for animals. It has been shown that the density of macroinvertebrates increases with particle size on sand in the Athabasca River, but it should be emphasized that the current regime in the main channel of that very large river is more uniform than would be expected in smaller streams, especially diversion channels whose discharge is not buffered by extensive muskegs. Also, the psammophilic fauna of the Athabasca River is characterized by forms which are highly adapted to constantly shifting sand. Thus, medium-sized sand particles are probably optimal for small streams since they represent a balance between maximum pore volume and minimum shifting by small fluctuations in current.

Growths of filamentous algae also supported a large percentage of the organisms found in the coarser sediment channels, especially on cobbles. The much greater surface area of stone exposed to light and current in the cobble channel supported nearly three times as many animals as did bank gravel, but these were mostly small species; the total weight of organisms on bank gravel was about nine times as great as on cobbles. If sphaeriid clams and elmid beetle larvae are excluded, the weight of organisms in bank gravel approached that in river gravel. The relative absence of these burrowing animals, as well as the smaller number of organisms and the smaller number of taxa, suggest that a conditioning period of more than 3 mo is necessary for the complete development of benthic communities on newly inundated gravels.

Observations throughout this study indicate that colonization of mineral substrates by benthic invertebrates is dependent upon the development of a microbial film on the surface of the stone and that this process is, to a large extent, light dependent. Thus, the more rapid colonization of cobbles, where a relatively greater proportion of the total inhabitable substrate is exposed to light, is exactly what would be expected. The ultimate density and variety of the fauna on more heterogeneous substrates, in this case gravels (which offer more potential surface area, a greater variety of potential microhabitats and more secure refuges for burrowing and negatively phototropic organisms), should be greater, but the development of microbial films is slower due to the reduced penetration of light into the more compact substrate.

On the basis of these results, it can be concluded that construction of diversion channels and the re-establishment of streams in mined areas, using limestone gravel for riffles and medium (overburden) sand for slow reaches, would probably provide for an optimal (i.e., nearly natural) biological productivity. Both materials are readily available in the study area and would require no processing before use, although the establishment of communities of organisms would be enhanced by the addition of organic detritus to the sand or gravel (Egglishaw 1964). Leaves and finely pulverized wood fragments would probably be best for this purpose, but the optimum ratio of detritus to inorganic material remain to be elucidated.

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6. APPENDIX

Table 42. Invertebrates collected from the AOSERP study area, 1976-77.

Invertebrate	Location[a]			
	M	S	AR	Other
PORIFERA				
Spongilla Lamarck	C	C		
CNIDARIA				
Hydra	C	C	I	
TURBELLARIA				
Alloecoela	I			
Tricladida				
Dugesia ?tigrina (Girard)	I			
NEMATODA	F	F	F	
NEMATOMORPHA	I	I		
HIRUDINOIDEA				
?Dina Blanchard	I	I		
Erpobdella punctata (Leidy)	C	I		Lakes
Glossiphonia complanata (Linnaeus)	I	C		
G. heteroclita	I			
Haemopis grandis (Verrill)	I	I		
Helobdella stagnalis (Linnaeus)	C	C		
Nephelopsis obscura Verrill		I		Lakes
Piscicola Blainville	I	I		
?Placobdella papillifera (Verrill)	I			
OLIGOCHAETA				
Lumbriculidae				
Lumbriculus variegatus (Müller)	C	I		

continued ...

Table 42. Continued.

Invertebrate	Location[a] M	S	AR	Other
Enchytraeidae	I	F	C	
Aeolosomatidae				
Aeolosoma sp. Ehrenberg			I	OT
Tubificidae	F,LA	F,LA	F,LA	
Limnodrilus ?claparedeanus Ratzel	I		I	
L. hoffmeisteri Claparède	I	I	I	
Peloscolex spp. Leidy	C	I		
Tubifex ?tubifex (Müller)		I		
Naididae				
Amphichaeta nr. *americana* Chen			F	
Arcteonais lomondi (Martin)	I			
Chaetogaster diaphanus (Guuithuisen)	C	I		
C. langi Bretscher	I			
C. limnaei von Baer				Lakes
Dero digitata (Müller)	I			
Nais behningi Michaelson	C,LA	F	F	
N. communis/variabilis	C	C		
N. pardalis Piquet	C			
N. pseudobtusa Piquet	I	I		
N. simplex Piquet	C	C	C	
Pristina breviseta Bourne	I	F		
P. foreli (Piquet)	I	I		
P. longiseta Ehrenberg	C			
Slavina appendiculata d'Udekem	C	C		
Specaria josinae (Vejdovsky)	I			
Stylaria lacustris (Linnaeus)	I	I		
Uncinais uncinata (Ørsted)	I	C		
Vejdovskyella comata (Vejdovsky)	I	I		

continued ...

Table 42. Continued.

Invertebrate	Location[a]			
	M	S	AR	Other
MOLLUSCA				
Pelecypoda				
Sphaeriidae				
Musculium Link	C	C		
Pisidium spp. Pfeiffer	F	F		
Sphaerium spp. Scopoli	C	F	C	
Unionidae				
Lampsilis sp. Rafinesque	I			
Gastropoda				
Amnicola ?limosa (Say)	I			
Ferrissia Walker	I			
Helisoma Swainson	I	I		
Gyraulus parvus (Say)	C	W		
Lymnaea spp. Lamarck	I,LA	I		Lakes
Physa Draparnaud	C	F		
Promenetus Baker	I			
Valvata ?lewisii Currier	C	C		
Bryozoa	I	I		
TARDIGRADA	I	I		
CRUSTACEA				
Copepoda	F,LA	F,LA	I	
Ostracoda	F	F	I	
Amphipoda				
Gammaridae				
Gammarus lacustris Sars	C,LA	I		
Talitridae				
Hyalella azteca (Saussure)	C,LA	C,LA		

continued ...

Table 42. Continued.

Invertebrate	Location^{a}			
	M	S	AR	Other
INSECTA				
Ephemeroptera				
Siphlonuridae				
Ameletus Eaton	I,LA	F	R	
Parameletus Bengtsson				OT
Siphlonurus Eaton	C	I		
S. alternatus (Say)	C	I		OT
Analetris eximia Edmunds			R	
Isonychia Eaton	I		I	
Metretopodidae				
Metretopus borealis Eaton	I,LA			
Siphloplecton basale (Walker)	I,LA	I		
Ametropodidae				
Ametropus neavei McDunnough		C,LA		
Baetidae				
Baetis Leach	C,LA	F,A	F	
Callibaetis coloradensis Banks	I	C		Lakes
Centroptilum Eaton	I	F		
Cloeon Leach	I			Lakes
C. implicatum McDunnough			I	
Pseudocloeon Klapalek	I	C		
Heptageniidae				
Epeorus (Iron) ?albertae (McDunnough)		I		
Heptagenia spp. Walsh	R,LA	F,A	C	
Rhithrogena Eaton	R	I	C	
Stenacron interpunctatum (Say)	I			
Stenonema vicarium (Walker)	R	C		
Pseudiron McDunnough			I	

continued ...

Table 42. Continued.

Invertebrate	Location[a]			
	M	S	AR	Other
Leptophlebiidae				
Leptophlebia cupida (Say)	F,A	C	I	
L. nebulosa (Walker)	F,A	C	I	
Paraleptophlebia Lestage	I	F	I	
Ephemerellidae				
Ephemerella margarita Needham	I	I		
E. simplex McDunnough	I,C	C		
E. spinifera Needham	I	F,A		
E. aurivillii Bengtsson	I			
E. inermis Eaton	I	C	F	OT
E. tibialis McDunnough	I	C		
Tricorythidae				
Tricorythodes ?minutus Traver	C,LA	C	I	
Caenidae				
Brachycercus Curtis	C,LA	I		
Caenis spp. Stephens	I	I	I	
Baetiscidae				
Baetisca ?columbiana Edmunds	I	I		
B. obesa (Say)	I	C		OT
Ephemeridae				
Ephemera cf. *simulans* Walker	I		I	Lakes
Hexagenia Walsh			I	
Odonata				
Anisoptera				
Aeshnidae				
Aeshna eremita Scudder	C	C		
A. nr. *interrupta* Walker	I	I		
A. ?umbrosa Walker	C	C		Lakes
Gomphidae				
Gomphus ?notatus Rambur			I	
Ophiogomphus colubrinus Selys	R,LA	C,A	C	OT

continued ...

Table 42. Continued.

	Location[a]			
Invertebrate	M	S	AR	Other
Corduliidae				
Cordulia shurtleffi Scudder	I			
Epitheca canis McLachlan	I			
Somatochlora minor Calvert	I	F		A,OT
Libellulidae				
Leucorrhinia borealis Hagen				Ponds
L. hudsonica (Selys)	I			
L. intacta Hagen				Ponds
Libellula julia Uhler				Ponds
L. quadrimaculata Linnaeus				Ponds
Zygoptera				
Agrionidae				
Agrion aequabile (Say)	I	I		OT
Coenagrion resolutum (Hagen)	I	I		
Enallagma boreale Selys	I			Lakes
Ishnura Charpentier	I			Ponds
Plecoptera				
Nemouridae				
Nemoura (Amphinemura) linda Ricker	I	I		OT
N. (Nemoura) arctica Esben-Peterson	R,LA	I		OT
N. (Shipsa) rotunda Claassen	C,LA	C,A	I	
N. (Zapada) cinctipes Banks	F			
Leuctridae				
Leuctra ?sara Classen	I	F		
Capniidae				
Capnia vernalis (Newport)	I	C	C	
Tainiopterygidae				
Oenopteryx fosketti (Newport)			C	
Taeniopteryx nivalis (Fitch)	C,LA	C		
T. parvula Banks	I			
Pteronarcidae				
Pteronarcella regularis (Hagen)		C	I	
Pteronarcys dorsata (Say)	R,A	C,A	C,LA	

continued ...

Table 42. Continued.

Invertebrate	Location[a]			
	M	S	AR	Other
Perlodidae				
Arcynopteryx Klapalek	R,LA	F		
Isogenus (Isogenoides) frontalis colubrinus (Hagen)	I	I	F,LA	
Isoperla nr. *fulva* Claassen	I			
I. ?fusca Needham & Claassen	R,LA	I		
I. longiseta Banks			F,A	
I. ?sordida (Banks)			I	
Chloroperlidae				
Hastaperla brevis (Banks)	R,LA	F	C,LA	
Perlidae				
Acroneuria abnormis (Newman)			C	
A. lycorias (Newman)	I			
Claassenia sabulosa Banks	R,?A	?C		
Megaloptera				
Sialidae				
Sialis Latreille	I	C		
Trichoptera				
Philopotamidae				
Wormaldia gabrielia (Banks)	R,LA	I		
Polycentropodidae				
Polycentropus cinereus Hagen	I			
P. flavus (Banks)	I		I	
P. remotus Banks			I	
Hydropsychidae				
Arctopsyche McLachlan	I	C	I	
Cheumatopsyche spp. Wallengren	C,LA	F	F,LA	
C. speciosa (Banks)			A	
Hydropsyche spp. Pictet	R,LA	F	C,LA	
H. bifida Banks	C	C	C	
H. slossonae Banks	C	C		

continued ...

Table 42. Continued.

Invertebrate	Location[a]			
	M	S	AR	Other
Rhyacophilidae				
Rhyacophila spp. Pictet	I	I		
Glossosomatidae				
Agapetus Curtis	I	I		
Glossosoma Curtis	R,LA	F		
Protoptila Banks	I	I		
Hydrotilidae				
Agraylea Curtis	I	I		
Dibusa Ross	I			
Hydroptila Dalman	I			
Mayatrichia Mosely	I	I		
Neotrichia Morton			I	
Ochrotrichia Mosely	I			
?*Orthotrichia* Eaton		I		
Oxyethira Eaton	R,LA	C		
Phryganeidae				
Agrypnia Curtis	I			Lakes
Banksiola crotchi Banks				Pond
Fabria Milne				Lakes
Phryganea Linnaeus	I			Lakes
Ptilostomis semifasciata (Say)	I	F		Lakes
Brachycentridae				
Brachycentrus Curtis	R,LA	F,A	F	
B. americanus (Banks)	R,LA	F,A		
Micrasema McLachlan	R,LA	F,A	I	

continued ...

Table 42. Continued.

Invertebrate	Location^a			
	M	S	AR	Other
Limnephilidae				
Anabolia bimaculata (Walker)	I			OT
Asynarchus McLachlan	C	I		OT,Lakes
Glyphopsyche irrorata (Fabricius)	I	C		
Grammotaulius Kolenati	I			
Hesperophylax Banks	I	C		
Limnephilus spp. Leach	C	F		
L. minusculus (Banks)	I			
Nemotaulius hostilis (Hagen)	I			Lakes
Onocosmoecus Banks		I		
Psychoglypha subborealis (Banks)	I	C		
Pycnopsyche Banks		R		
Lepidostomatidae				
Lepidostoma Rambur	R,LA	F,A	I	
Molannidae				
Molanna Curtis				Lakes
Helicopsychidae				
Helicopsyche borealis Banks	R			
Leptoceridae				
Ceraclea annulicornis (Stephens)	I	C		
C. tarsipunctata (Vorhies)	I	C	R	
Nectopsyche Müller	I			
Oecetis avara Banks	I	I	C	
Triaenodes McLachlan			I	
Lepidoptera				
Pyralidae				
Nymphula Schrank			I	

continued ...

Table 42. Continued.

Invertebrate	Location[a]			
	M	S	AR	Other
Hemiptera				
Corixidae				
Arctocorisa sutilis (Uhler)				Lakes
Callicorixa audeni Hungerford		F,A	I,LA	Lakes
C. alaskensis (Hunderford)				Lakes
Cenocorixa dakotensis (Hungerford)				Lakes
Hesperocorixa atopodonta (Hungerford)	C	C		
H. michiganensis (Hungerford)	I	F		
H. minorella (Hungerford)		I		
Sigara alternata (Say)	I,LA	I,LA		
S. bicoloripennis (Walley)	I	C		Lakes
S. conocephala (Hungerford)	I	C	I,LA	Lakes
S. decoratella (Hungerford)	I	C		Lakes
S. fallenoidea (Hungerford)				Lakes
S. grossolineata Hungerford		I		
S. lineata (Forster)			I	
S. mullettensis (Hungerford)	I	I		
S. penniensis (Hungerford)	I	I		Lakes
S. solensis (Hungerford)	I,LA	C	I	Lakes
S. trilineata (Provancher)			I	
S. washingtonensis Hungerford		I		
Trichocorixa borealis Sailer				L.Athabasca
T. naias (Kirkaldy & Bueno)	I			Lakes
T. verticalis interiores Sailer		I		
Notonectidae				
Notonecta borealis Bueno & Hussey				Lakes
N. kirbyi Hungerford				Lakes
N. undulata Say				Lakes

continued ...

Table 42. Continued.

	Location[a]			
Invertebrate	M	S	AR	Other
Coleoptera				
Haliplidae				
Brychius Thomson	I	I,LA		
Haliplus	I	C		Lakes
Dytiscidae				
Agabus spp. Leach	I	I		
A. seriatus (Say)	I			OT
Carrhydrus crassipes Fall	I			
Deronectes Sharp		I		
Dytiscus dauricus Gebler				OT
D. harrissi Kirby	I			
Hydaticus Leach		I		
Hydroporus Clairville		I		
Ilybius Erichson	I			
Neoscutopterus Balfour-Brown		I		
Gyrinidae				
Gyrinus affinis Aube	C,LA			
G. maculiventris LeConte	I	I		
G. minutus Fabricius	I			
G. ?opacus Sahlberg		C		
G. ?pectoralis LeConte	I			
Hydrophilidae	C			
Elmidae				
Dubiraphia robusta Hilsenhoff	I			
Optioservus fastiditus (LeConte)	R,LA	I	I	

continued ...

Table 42. Continued.

Invertebrate	Location[a]			
	M	S	AR	Other
Diptera				
Tipulidae				
Antocha Osten Sacken	I	I		
Dicranota Zetterstedt	R,LA	F		
Eriocera Macquart	R,LA	C	I	OT
Holorusia Loew	I			
Prionocera Loew		I		
Tipula Linnaeus	I	I		
Psychodidae				
Pericoma Walker		I		
Telmatoscopus Eaton	I	I		
Dixidae				
Paradixa Tonnoir	I			
Chaoboridae				
Chaoborus Lichtenstein				Ponds
Simuliidae				
Simulium arcticum Malloch	I		F,A	
S. prob. *aureum* Fries	C			
S. decorum Walker	C	C		
S. euryadminiculum Davies	I			
S. prob. *tuberosum* Lundström				OT
S. tuberosum-complex	C,A	C,A	C,A	
S. venustum-complex		C	C	
S. vittatum Zetterstedt				OT
Ceratopogonidae	F	F	C	
Atrichopogon Kieffer	I			
Chironomidae				
Podonominae				
Trichotanypus posticalis (Lundbeck)	R			

continued ...

Table 42. Continued.

Invertebrate	Location[a]			
	M	S	AR	Other
Tanypodinae				
Ablabesmyia spp. Johannsen	F	F		
Conchapelopia Fittkau	I	C		
Labrundinia Fittkau	I	I		
Larsia Fittkau	C	C	I	
Nilotanypus Kieffer	R,C	C	I	
Paramerina Fittkau	I	C		
Procladius Skuse	F	F	I,LA	
Rheopelopia Fittkau	I			
Thienemannimyia Fittkau	I			
Thienemannimyia-gp.	C	F	F	
Chironominae - Chironomini				
Beckiella tethys (Townes)			F	
Chernovskiia orbicus (Townes)			F	
Chironomus annularis-gp.	I			
C. fluviatilis-gp.	C	I	I,LA	
C. cf. *decorus* (Johannsen)	C			
C. plumosus-gp.	I	I		
C. salinarius-gp.	I	I	I	OT
C. thummi-gp.	I,LA			
Cladopelma Kieffer	C	I		
Cryptochironomus Kieffer	C	F	F	
"Cryptochironomus" rolli Kirpitshenko			C	
Cryptocladopelma Lenz	I	I		
Cryptotendipes Lenz	I	I	I	
Cyphomella cf. *gibbera* Saether			C,LA	
Demicryptochironomus Lenz			I	OT
Dicrotenipes cf. *fumidus* (Johannsen)	C	C		
D. cf. *modestus* (Say)	I			
D. cf. *neomodestus* (Malloch)	I			

continued ...

Table 42. Continued.

Invertebrate	Location[a]			
	M	S	AR	Other
D. cf. *nervosus* (Staeger)	I	I		L.Athabasca
Endochironomus cf. *subtendens* (Townes)	C	I	I	
Glyptotendipes Kieffer	I	I		
Kiefferulus Goetghebuer		I		Ponds
Microtendipes cf. *pedellus* (DeGeer)	C	C	I	
Pagastiella				
Parachironomus Lenz	C	I		
Paracladopelma spp. Harnisch	I	I	C,A	
Paralauterbourniella Lenz	I	I	I	
Paratendipes Kieffer	I	C	C	
Phaenopsectra Kieffer	C	F	I	
Polypedilum Kieffer	C	F	F	
P. breviantennatum-gp.	I	F	F,A	
P. fallax-gp.	I	C	I	
P. scalaenum-gp.			F	
Robackia claviger (Townes)			F,A	
R. demeijerei (Kruseman)			F,A	
Stenochironomus Kieffer			I	
Stictochironomus spp. Kieffer	C	F	C	
Xenochironomus xenolabis (Kieffer)	I			
Tanytarsini				
Cladotanytarsus Kieffer	C	F	C	
Constempellina Brundin		I		
Micropsectra Kieffer	F,A	F,A	C	
Paratanytarsus Kieffer	C	I		
Rheotanytarsus (Bause)	C,A	F,A	F	
Stempellina spp. Bause	C,LA	F		
Tanytarsus van der Wulp	F,A	F,A	C	
Zavrelia Kieffer	C	F	C	

continued ...

Table 42. Continued.

Invertebrate	Location[a]			
	M	S	AR	Other
Diamesinae				
Diamesa (Meigen)	I	I		
Monodiamesa cf. *tuberculata* Saether			C	
Potthastia cf. *gaedi*	I			
P. longimanus-type	I	C	I	
Protanypus Kieffer	I			
Pseudodiamesa Goetghebeur	I		I	
Orthocladiinae				
Acricotopus cf. *senex* (Johannsen)	C	I		
Brillia Kieffer	C	C	C	
Cardiocladius Kieffer	C	C		
Corynoneura Winnertz	F	F	I	
Cricotopus bicinctus (Meigen)	C,A	C,A	I	Ponds
C. cylindraceus-gp.	I	I	I	
C. Juscus-gp.		I	I	
C. tremulus-gp.	C,A	C,A		
C. nr. *curtus* Hirvenoja	I	I		OT
C. cf. *triannulatus* Goetghebuer		C		
C. trifascia-gp.	I			
C. nostocicola wirth	I	C		
C. festivellus-gp.				Pond
C. sylvestris-gp.				L.Athabasca
C. cf. *laetus* Hirvenoja		I		
Diplocladius cf. *cultriger* (Kieffer)	C	F		
Eukiefferiella spp. Thienemann	C	F,A	C	
E. cf. *brevicalcar* Kieffer	I	I		
E. cf. *claripennis* (Lundbeck)	I,LA	C	I	
Eurycnemus van der Wulp	I			
Heterotrissocladius cf. *latilaminus* Saether	C	F	I	
Krenosmittia Thienemann	I	I		

continued ...

Table 42. Continued.

Invertebrate	Location[a] M	S	AR	Other
Limnophyes Eaton	I	I	I	
Metriocnemus van der Wulp	I			
Nanocladius cf. *balticus* (Palmén)		I		
N. cf. *distinctus* (Malloch)	C			
N. cf. *rectinervis* (Kieffer)		F	C	
Orthocladius spp. (van der Wulp)	I	F		
Parakiefferiella spp. (Thienemann)	C	F	I	
Parametriocnemus spp. Goetghebuer	C	F		
P. cf. *graminicola* (Lundbeck)		I		
P. cf. *lundbecki* (Johannsen)	I	C		
Paraphanocladius Thienemann		C		
Paratrichocladius Thienemann	I,LA			
Psectrocladius spp. (Kieffer)	C	C		
P. cf. *simulans* (Johannsen)	I	I		OT
Pseudosmittia (Goetghebuer)	I			
Rheocricotopus (Thienemann & Harnisch)	I,LA	C	I	
R. nr. *kenorensis* Saether	I,LA			
Synorthocladius Thienemann	I	F	I	
Thienemanniella Kieffer	C	F	I	
?Genus acutilabis Pankratova		C		
Orthocladiinae A	I,LA	C		
Orthocladiinae B			F,A	OT
Orthocladiinae D		I	I	
Stratiomyidae				
Stratiomyia Geoffroy		I		
Rhagionidae				
Atherix pachypus Bigot	R,LA	C	I	
Tabanidae				
Chrysops Meigen	I	C		
Dolichopodidae		C	I	

continued ...

Table 42. Concluded.

Invertebrate	Location[a]			
	M	S	AR	Other
Empididae				
Chelifera Macquart	C	C	I	
Hemerodromia Meigen	C	F	C	
H. rogatoris Coquillett	C			
Rhamphomyia (Megacyttarus) Meigen	LA	LA		
Wiedemannia Zetterstedt		C	I	
Syrphidae				
Helophilus Meigen	I	I		
Anthomyiidae				
Limnophora Robineau - Desvoidy		I		
Ephydridae				
Psilopa Fallen	I	I		OT

[a] Location codes:

M = Muskeg River

S = Steepbank River

AR = Athabasca River

F = frequently collected

C = common

I = infrequent

A = abundant

LA = locally abundant

R = restricted to lower reaches

OT = other tributary streams

6.1 SPECIES LIST OF ALGAE FOUND ON OILED AND CONTROL BRICKS FROM JULY TO DECEMBER 1977, MUSKEG RIVER.

Cyanophyta

Aphanocapsa sp.

A. pulchra Kuetzing Rabenhorst

A. elachista West & West

Calothrix epiphytica West & West

C. breviarticulata West & West

Choemosiphon incrustans Grunou

Chrooccoccus turgids (Keutzing) Naegeli

Lyngbya sp.

L. taylorii Dronet & Strickland

L. aergineo caerulea (Kuetzing) Gomont

L. epiphytics Hieronymus

Phormidium sp.

P. autumnale (C.A. Agardh) Gomont

P. tenue (Menegh) Gomont

Oscillatoria tenuis var. *tergestina* Keutzing

O. lacustris (Klel) Geitler

Microcystis aeruginosa Keutzing

Schizothrix tinctoria Gomont

Spirulina laxa G.M. Smith

Rivularia haematites (D.C.) C.A. Agardh

Chlorophyta

Ankistrodesmus sp.

Bulbachaete sp.

Cladophora glomerate (L.) Kuetzing

Euglena gracilis Klebs

Ulothrix zonata (Weber & Mohr) Kuetz

Draparnaldia plumosa (Vauch.) C.A. Agardh

D. oacuta (C.A. Agardh) Kuetzing

Rhodophyta

Audouinella chalybea (Lyngb.) Fries

A. pygmaea Kuetzing

Bactrachospermum vagum (Roth) C.A. Agardh
Achnanthes minutissima Kuetzing
A. lanceolata (Breb.) Grunow
A. exigua Grunow
Amphora ovalis Kuetzing
Amphipleura pellucida Kuetzing
Caloneis bacillum Grunow
Cocconeis placentula Ehrenberg
Cymbella ventricosa Kuetzing
C. caespitosa (Kuetzing) Brun
Cymatopleura solea (Breb.) W. Smith
Diatoma elongatum Agardh
D. vulgare Bory
Epithemia zebera Ehrenberg
E. sorex Kuetzing
Fragillaria construens (E) Grun
F. capucina Desmazieres
Gomphonema acuminatum Ehrenberg
G. olivaceum (Lyngb.) Kuetzing
G. parvulum Kutz
G. constrictum (Ehrenberg)
G. gracile E. Cleve
Melosira sp.
Navicula pygmeae Kuetzing
N. nuralis Grunow
N. rhyncocephala Kuetzing
N. viridula Kuetzing
N. cryptocephala Kuetzing
N. halophila Grunow
N. anglica Ralfs
N. radiosa Kuetzing
Nitzschia sublenearis (Hustedt)
N. acicularis W. Smith
N. amphilira Grunow

N. fronticola Grunow

N. perminuta Grunow

N. sp.

Rhoicosphenia curvata Keutzing Grunow

Rhapoldiagibba (Ehr.) O. Mull

Stauroneis legumen Ehrenberg

Synedia ulna Nitzsch Ehrenber

S. rumpens Keutzing

S. parasitica W.M. Smith

S. acus Keutzing

Additional algal species on the light/dark oil experiment from Section 3.2.

Cyanophyta

Anabaena sp.

Phormidium favosum (Bory) Gomont

Gleotrichia longiarticulata G.S. West

Bacillariophyta

Tabellaria flocculosa (Roth) Keutzing

Synedra pulchella var. *minuta* (Ralfs) Keutzing

Cymbella gracilis (Rabenh)

Navicula bacillum Ehrenberg

Cyclotella sp.

Navicula oblonga-subcapitata Keutzing

N. radiosa Keutzing

Cocconeis placentula var. *lineata* (Ehrenberg) Cleve

Chlorophyta

Stigeoclonium pachydermum Prescott

7. AOSERP RESEARCH REPORTS

1.		AOSERP First Annual Report, 1975
2.	AF 4.1.1	Walleye and Goldeye Fisheries Investigations in the Peace-Athabasca Delta--1975
3.	HE 1.1.1	Structure of a Traditional Baseline Data System
4.	VE 2.2	A Preliminary Vegetation Survey of the Alberta Oil Sands Environmental Research Program Study Area
5.	HY 3.1	The Evaluation of Wastewaters from an Oil Sand Extraction Plant
6.		Housing for the North--The Stackwall System
7.	AF 3.1.1	A Synopsis of the Physical and Biological Limnology and Fisheries Programs within the Alberta Oil Sands Area
8.	AF 1.2.1	The Impact of Saline Waters upon Freshwater Biota (A Literature Review and Bibliography)
9.	ME 3.3	Preliminary Investigations into the Magnitude of Fog Occurrence and Associated Problems in the Oil Sands Area
10.	HE 2.1	Development of a Research Design Related to Archaeological Studies in the Athabasca Oil Sands Area
11.	AF 2.2.1	Life Cycles of Some Common Aquatic Insects of the Athabasca River, Alberta
12.	ME 1.7	Very High Resolution Meteorological Satellite Study of Oil Sands Weather: "A Feasibility Study"
13.	ME 2.3.1	Plume Dispersion Measurements from an Oil Sands Extraction Plant, March 1976
14.		
15.	ME 3.4	A Climatology of Low Level Air Trajectories in the Alberta Oil Sands Area
16.	ME 1.6	The Feasibility of a Weather Radar near Fort McMurray, Alberta
17.	AF 2.1.1	A Survey of Baseline Levels of Contaminants in Aquatic Biota of the AOSERP Study Area
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